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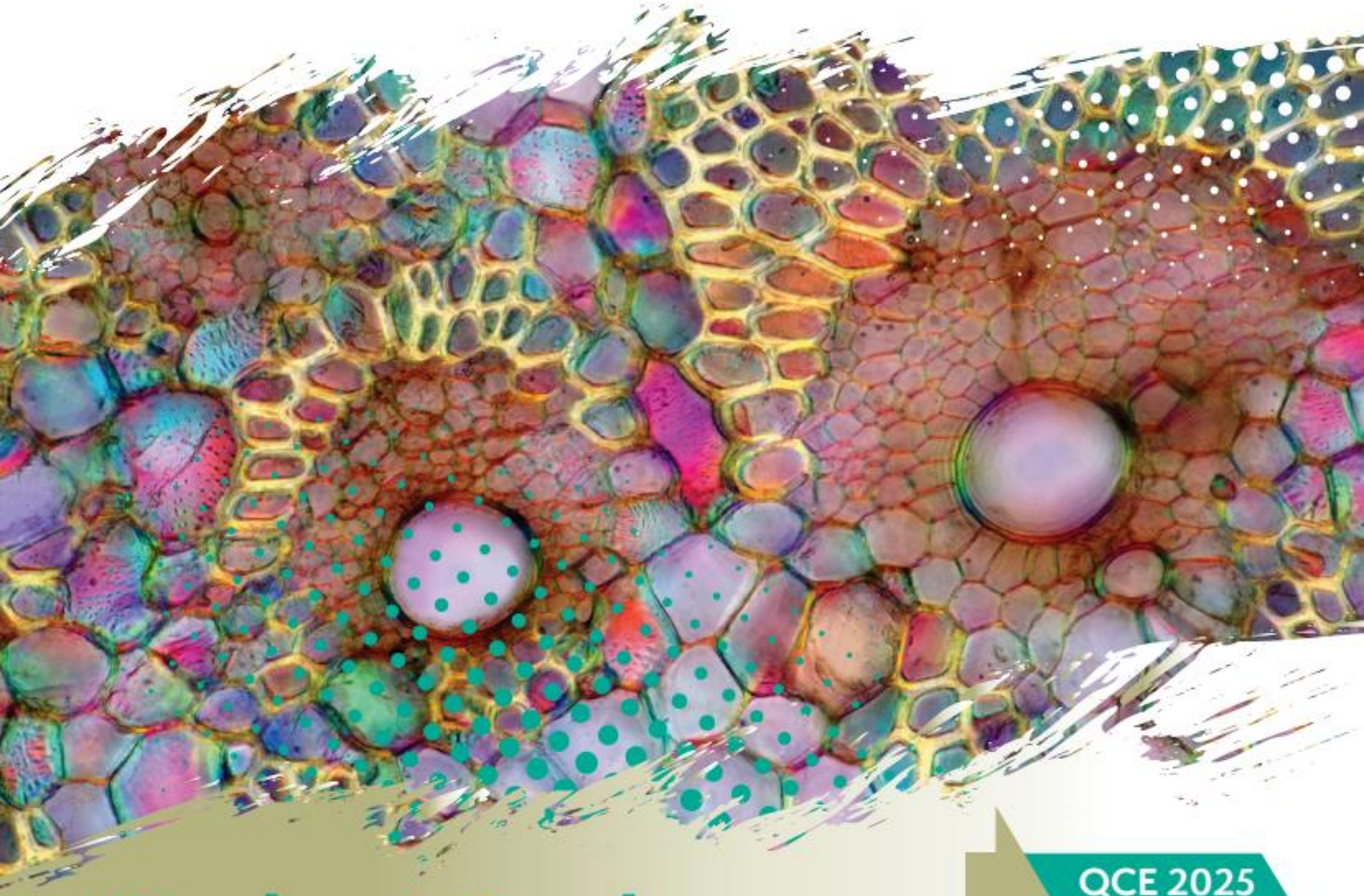
2nd EDITION



Student Book

QCE 2025 Biology SYLLABUS

PEARSON

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QUEENSLAND

UNITS 1 & 2

STUDENT BOOK

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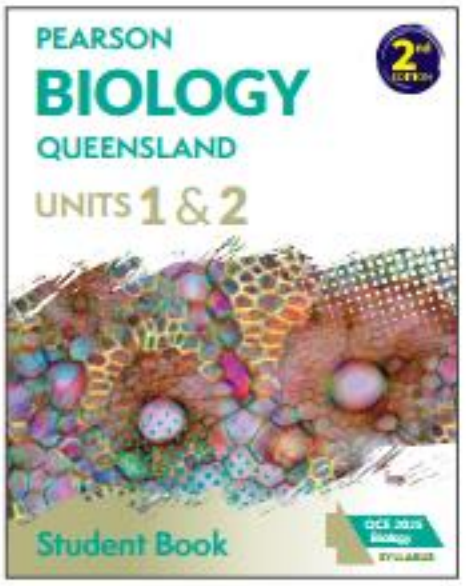

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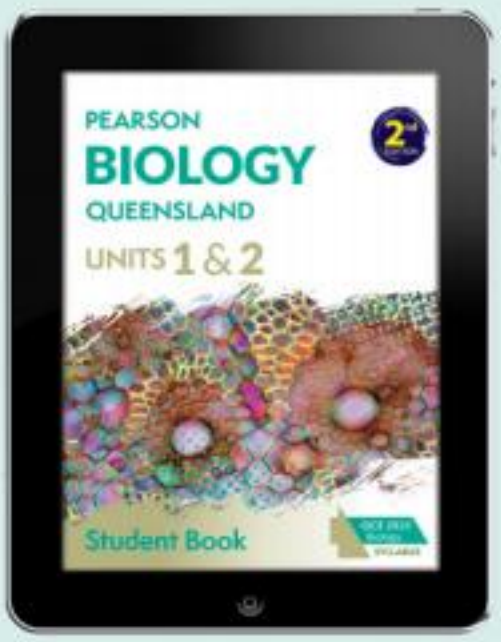

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How to use this book

PEARSON BIOLOGY QUEENSLAND STUDENT BOOK UNITS 1 & 2 2ND EDITION

Pearson Biology Queensland Student Book Units 1 & 2 2nd edition has been written to the 2025 QCAA Biology Syllabus. The book includes an additional Skills and Assessment Toolkit online as well as a depth of additional resources, to support every aspect of teaching and learning. Explore how to use this book below.

Design

The best-practice literacy and instructional design supports all learners.

A simple to navigate, predictable design enables ease of use. The high-quality, relevant photos and illustrations assist students' understanding of the concepts.

Chapter 1

The Skills and Assessment Toolkit provides support across the Science Inquiry Skills and Assessment requirements. The material is presented as a handy reference to enable confident, complete coverage of the syllabus across the course. Reworked and fully updated to meet the evolved requirements of the revised QCE 2025 Syllabus and to respond to the needs of teachers and students.

Chapter opener

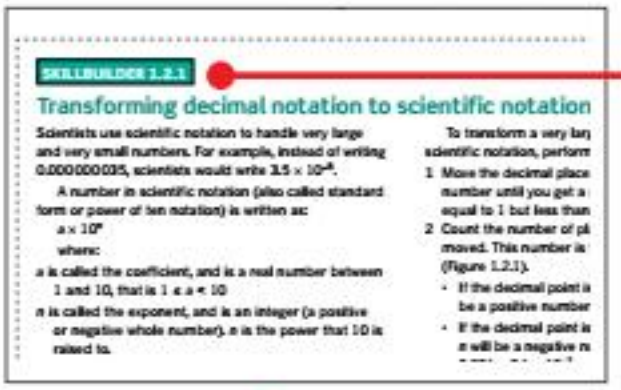
The syllabus subject matter addressed in each chapter is clearly listed, and mapped to the modules in which the content is addressed.

Module opener

Module openers outline the key concepts and skills to be developed and link to the syllabus subject matter listed in the Chapter opener.

Skillbuilders

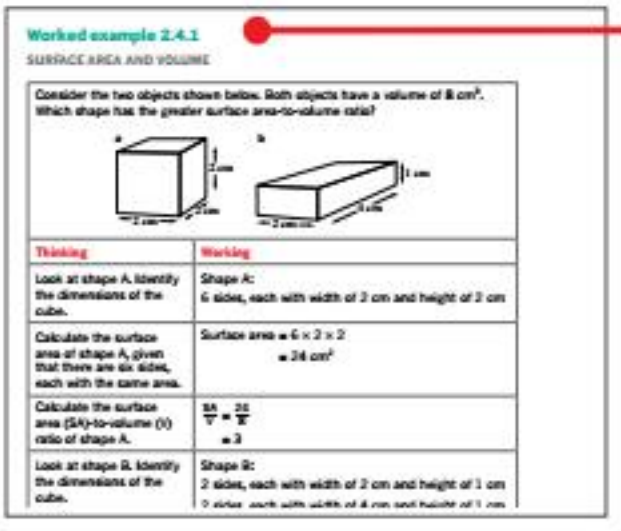
Skillbuilders outline a method or technique. Each is instructive and self-contained. Skillbuilders provide students with the steps needed to apply the skill, supporting development of key science inquiry skills.



Worked examples

Worked examples use sequential steps of thinking and working to model a best-practice approach to scaffolding student development of subject matter. Each Worked example is followed by a Try yourself task where students apply their learning to a mirrored problem.

Fully worked solutions to all Try yourself problems are available from *Pearson Biology Queensland Units 1 & 2 2nd edition* eBook.



> Peptidoglycan is a macromolecule made of protein and carbohydrate, found only in bacteria.

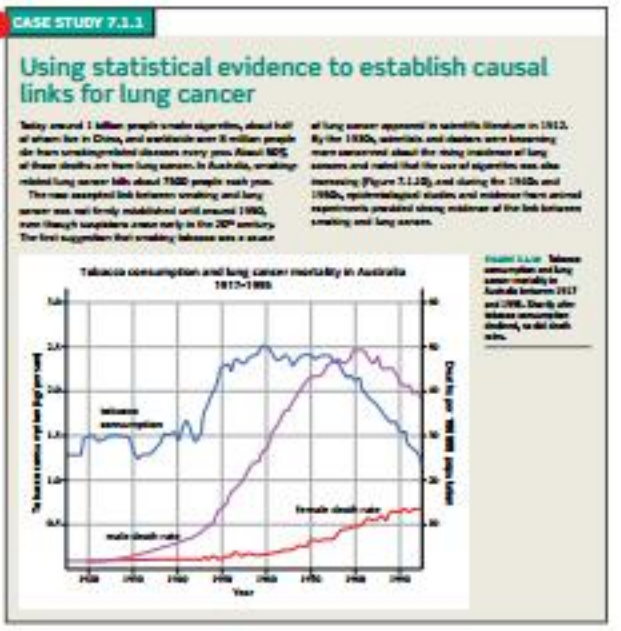
Highlight box

Highlight boxes focus on important information such as key definitions, formulas and salient points.

How to use this book

Case studies

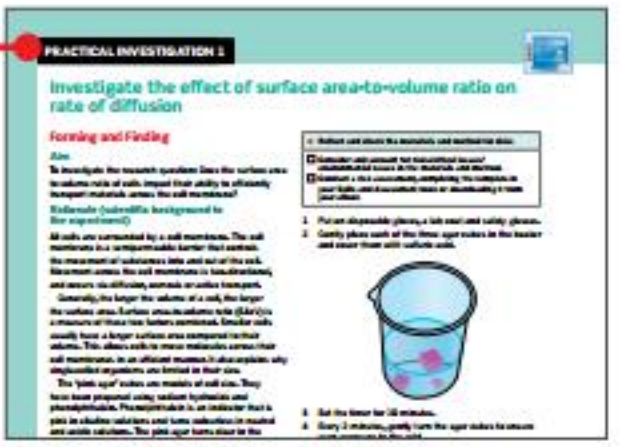
Case studies provide opportunities to engage with current applications and research in biology and address essential syllabus objectives beyond typical learning and understanding conventions. Case studies develop skills in analysing, interpreting, evaluating, decision-making and predicting. Skills are modelled for students in the Case studies and then learning is applied in the Case study review.


CASE STUDY 7.1.1

Using statistical evidence to establish causal links for lung cancer

Practical manual

Practical investigations offer students the opportunity to develop, practise and refine their Science inquiry skills. These activities involve gathering primary data to analyse and respond to scaffolded or open-ended science tasks, supporting deeper understanding and proficiency in scientific methods. Additionally, these tasks allow teachers to formatively assess students' science inquiry skills and set students up well with the insights for Student experiment IA2, pertinent to Assessment objectives 1 to 6. All Practical investigations have been trialled and tested to ensure they can be safely performed and yield effective results. Full risk assessments and additional support are provided in the eBook.


PRACTICAL INVESTIGATION 1

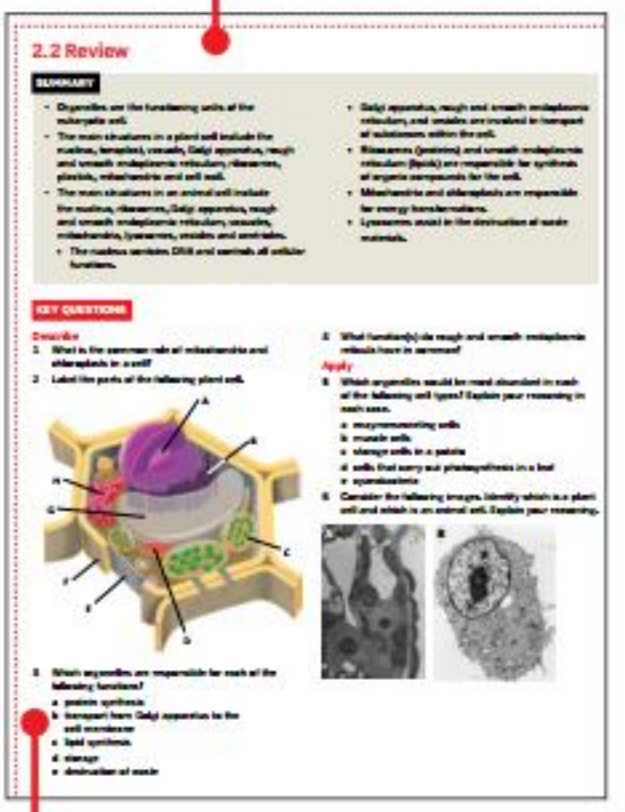
Investigate the effect of surface area-to-volume ratio on rate of diffusion

Science as a human endeavour

Science as a human endeavour (SHE) features provide stimulus for the Research investigation. The SHE features are structured to develop the foundational skills relevant to supporting the relevant Science inquiry skills through the lens of Science as a human endeavour, providing a suitable foundation from which students can launch into Research investigation IA3 and draw upon building proficiency for Assessment objectives 1, 2 and 3.

Module summary

Each module concludes with a summary to help students consolidate the key points and concepts.


2.2 Review

Module review

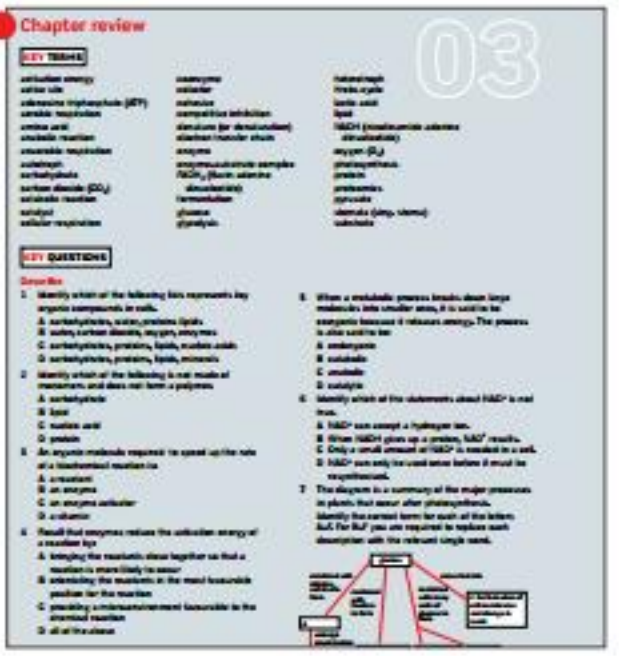
Key questions are provided to test students' understanding of concepts of the module. Questions are presented under the three Assessment objectives of Describe, Apply and Analyse. This structure allows students to demonstrate their progress in developing an understanding of the science subject matter.

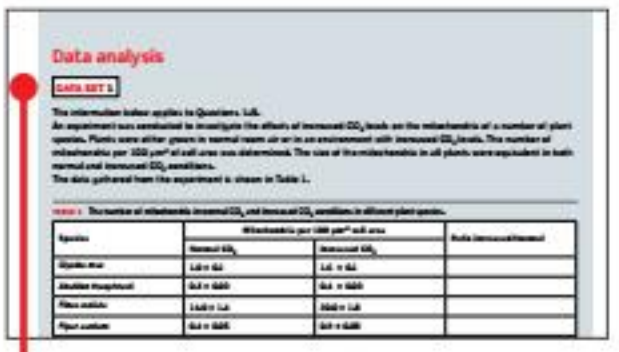
SCIENCE AS A HUMAN ENDEAVOUR 1

Organising complex biological systems

Chapter review

Each chapter finishes with a list of key terms covered in the chapter, and a set of multiple choice and short response questions, reflecting the style and experience of the External Assessment (EA). These questions are organised under the four Assessment objectives for the subject matter: Describe, Apply, Analyse, and Interpret.



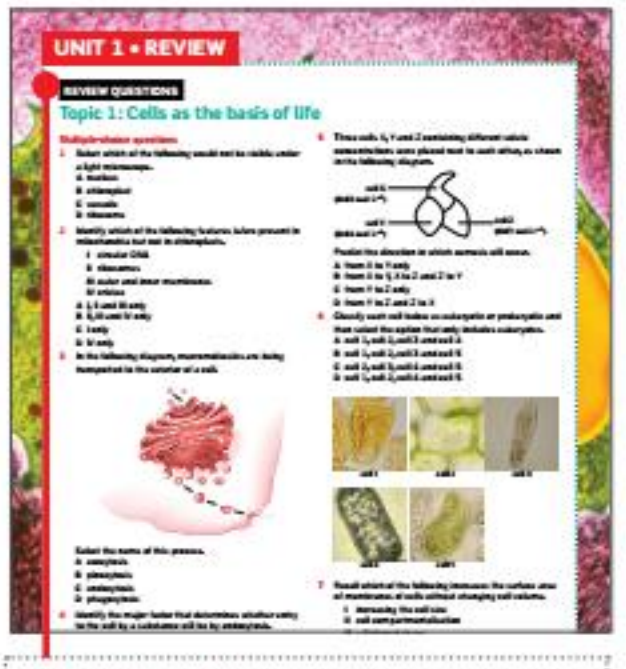


Data analysis

Data analysis tasks at the end of each Chapter review, provide an opportunity to respond to questions that use a range of qualitative and quantitative data relevant to that subject matter. The Data analysis tasks are structured to reflect the approach taken in the Data Test IA1 and align with the Assessment objectives 2, 3 and 4.

Glossary

Key terms are shown in **bold** throughout the Student Book and are listed at the end of each chapter. A comprehensive glossary at the end of the book defines all the key terms and aligns with the syllabus context.



Unit review

Each unit finishes with a comprehensive set of exam-style questions, including multiple choice, short answer and extended response. These review tasks assist students to draw together their knowledge and understanding of the whole unit.

Answers

Comprehensive answers and fully worked solutions for all Module review tasks, Try yourself, Science as a human endeavour, Case studies, Chapter reviews and Unit reviews are provided via the eBook.

Icons

Go To icons make important links to relevant content within the student books in the course. The Go To icons indicate:

GO TO ➤

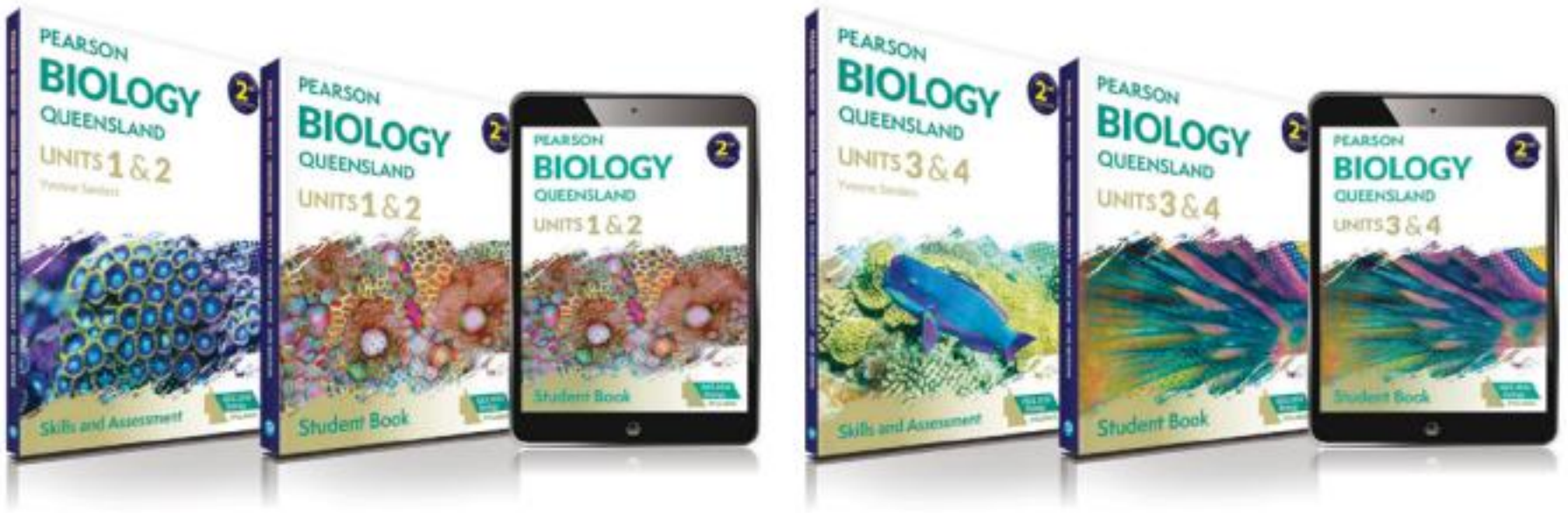
- when a Practical investigation would be suitable to engage with
- where support is available for skills and assessment tasks in Chapter 1 of your eBook.

Every Practical investigation is supported by a complementary **SPARKlab** alternative practical.

Pearson

Pearson Biology Queensland Units 1 & 2 2nd edition

Trusted throughout Queensland to provide high quality support for implementation of the QCE Biology 2025 Syllabus, **Pearson Biology Queensland 2nd edition** has been refined to include what is needed to cover the QCE outcomes.



Student book

- The expert author team has delivered the same high-quality content you can trust, in the streamlined new edition for Queensland science classrooms.
- New, enhanced, and updated features that make the new second edition Student Books stand out:
  - New Science as a Human Endeavour sections
  - New Data Analysis tasks at the end of each Chapter review
  - Enhanced and updated Skills and Assessment Toolkit
  - Enhanced question sets
  - Updated Practical Manual of Practical Investigations
  - Updated Unit reviews and glossary.

Skills and assessment book

- The Skills and Assessment Book gives students the edge in preparing for all forms of assessment.
- It has been updated and refined to respond to the revised syllabus, and specifically provide opportunities to consolidate, develop and apply subject matter and Science inquiry skills. This resource features:
  - An updated Toolkit
  - Key knowledge summaries
  - Worksheets
  - Practical investigations and guidance
  - Assessment practice
  - Exam-style topic review sets.

eBook

Learner eBook access includes:

- Student book content
- Access to digital resources
- Fully worked solutions.

Teacher eBook access includes:

- Access to everything learners have access to
- Practical investigation expected results, common mistakes, suggested answers and full safety notes and risk assessments
- Teaching, learning and assessment programs
- Syllabus mapping documents.

Find out more and explore the series at

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CHAPTER

01 Biology skills and assessment toolkit

This chapter provides important information and support in the study of the QCAA Biology General Senior Syllabus for Units 1 to 4.

The Biology skills and assessment toolkit is designed to be used as a reference tool. It should be consulted on a need-to-know basis, where relevant, during this course of study. It is not intended that this chapter be worked through as a whole.

Focus

This chapter focuses on providing support and guidance in:

- **Development and application of scientific skills**
  - Mathematical and statistical processes used in Biology
  - SI units
  - Visual representations
  - Graphical representations
  - Measurement errors and uncertainty
  - Scientific writing conventions
- **Completing the assessment tasks**
  - Data test
    - Preparation in skill development
  - Student experiment
    - Forming a relevant research question
    - Justifying experiment modifications
    - Developing a methodology
    - Finding sufficient relevant data
    - Interpreting and analysing evidence
    - Evaluating experimental process
    - Scientific writing and representations
    - Referencing sources
  - Research investigation
    - Developing a research question from a claim
    - Collecting relevant and sufficient evidence to answer research question
    - Identifying and analysing trends, patterns, relationships in the evidence
    - Evaluating the quality and limitations of the evidence
    - Evaluating the research process
    - Scientific writing and representations
    - Referencing sources

Assessment

In Units 1 and 2, students are required to complete at least two and no more than four assessment tasks developed by the teacher. At least one assessment task must be completed per unit. In preparation for Biology Units 3 and 4, teachers may choose to mirror some of the assessment requirements of Units 3 and 4 in assessing Units 1 and 2.

The support material in this chapter is based on requirements in Units 3 and 4 and relates to the following assessments:

- Data test (Internal Assessment IA1)
- Student experiment (Internal Assessment IA2)
- Research investigation (Internal Assessment IA3)

Chapter organisation

The chapter is arranged in three parts:

- Part A: Working scientifically
- Part B: Student experiment
- Part C: Research investigation.

An outline of these parts is provided below.

QCAA Biology Syllabus objectives

- describe ideas and findings
- apply understanding
- analyse data
- interpret evidence
- evaluate conclusions, claims and processes
- investigate phenomena

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Part A: Working scientifically (IA1)

Part A focuses on basic mathematical skills and their applications. It features many worked examples and opportunities to apply these examples yourself. These skills are assessed, directly and indirectly, in assessment tasks and by analysing data in practical investigations. Part A focuses on skills for the data test (IA1) and experiments. These skills are useful when completing sections of the student experiment, research investigation and the examination. Part A also provides support in writing scientifically, avoiding plagiarism, referencing sources and in-text citations to assist a student's OR student experiment and research investigation.

Refer to the following outline of Part A: Working scientifically, to learn, revise and practise the skills in the areas in which you need help.

Module	Look here for	eBook page
1.1 Biological science	• areas of biology • the work of scientific research	e7
1.2 Orders of magnitude and estimation	• scientific notation • transforming decimal notations to scientific notation • converting between scientific notation and scientific units, orders of magnitude and prefixes • SI prefixes • examples of calculations	e12
1.3 Mathematical basics for biology	• examples of maths used in biology • analysing straight line graphs	e18
1.4 Units	• measurement and units • correct use of unit symbols • SI units • unit conversions	e26
1.5 Measuring precision and accuracy using uncertainties and percentage error	• explanations of the terms: uncertainty, error, accuracy, precision • causes of errors • calculations with uncertainty and error • writing measurements with uncertainty and error values • writing measurements and calculations to the correct significant figure • determining the absolute uncertainty of a value • converting between absolute and percentage uncertainties • calculating a value along with the associated uncertainty • calculating percentage error	e31

GO TO ➤ your eBook to access Chapter 1 Biology Skills and Assessment Toolkit

1.6 Tables and graphing	• arranging and recording data in summary tables
	• converting data from tables to graphs
	• types of graphs
	  - pie charts
	  - line graphs
	  - bar graphs
	  - scatterplots (with trendline and R2)
	  - column graphs (with error bars)
	• outliers and how they are represented on graphs
	• representing missing data on graphs
		e49

1.7 Data analysis	• calculating mean, median, mode, range • correlational investigations: regression analysis, Pearson's correlation coefficient, Spearman's rank	e60
1.8 Scientific writing	• writing scientifically • developing scientific representations • maintaining a science journal • referencing sources • in-text citations • plagiarism • referencing AI sources • multimodal presentations	e75

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Part B: Student experiment (IA2)

Part B focuses on the student experiment. This internal assessment task requires students to follow the full scientific method over an extended and defined period. Students develop their own research question to investigate, based on an identified experiment related to Unit 3 subject matter (or as relevant for Unit 1).

Part B supports all aspects of the student experiment. The QCAA objectives and instrument-specific marking guide (ISMG) for the assessment are explained.

Part B provides examples of how to **modify** (**extend**, **refine** or **redirect**) the initial experiment to write an experiment research question. Included is a step-by-step guide to evaluate the quality of the question and explanations of scientific methodology. Part B also covers data types, data collection and analysis of data to draw valid conclusions. It includes how to evaluate the quality of data, including its reliability and validity, and how to link these to logical improvements and extensions. Also provided is support in writing your scientific report, covering writing conventions and referencing sources.

Refer to the following outline of Part B: Student experiment, to learn, revise and practise your skills in the areas in which you need help.

Module	Look here for	eBook page
1.9 Forming	• establishing background information • developing a rationale • defining variables • developing the research question • evaluating the research question • justifying the question modification • putting it all together–sample report	e90
1.10 Finding	• determining the risk assessment • managing risk • considering environmental and ethical issues • collecting sufficient and relevant raw data • collecting data electronically • putting it all together–sample report	e101
1.11 Analysing	• presenting the data • processing data using mathematical techniques • identifying patterns, relationships, trends in data • interpreting results • identifying uncertainty and limitations in the evidence • putting it all together–sample report	e110
1.12 Interpreting and evaluating	• drawing a conclusion that is linked to the research question • discussing reliability and validity • identify improvements and extensions to the research • putting it all together–sample report	e116

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Part C: Research investigation (IA3)

Part C focuses on the research investigation. This internal assessment task requires students to gather secondary evidence on a research question. It is completed over an extended and defined period. Students develop their own research question to investigate, based on a claim related to Unit 4 subject matter (or as relevant for Unit 2).

Part C supports you through all aspects of the research investigation. The QCAA objectives and instrument-specific marking guide (ISMG) for the assessment are explained.

Part C assists with writing your research question. It considers examples of claims and how to analyse claims for context, and guides you in developing your own research question. You will be guided in writing a specific and relevant research question that encompasses a small sub-section of the claim that is linked to Biology syllabus concepts. You will consider the information on locating and evaluating suitable secondary sources for the research, including identifying relationships in data and assessing validity. You will learn how to formulate a justified conclusion and describe improvements and extensions. There is support for writing your scientific report including using the scientific writing style and structuring your report.

Refer to the following outline of Part C: Research investigation, to learn, revise and practise your skills in the areas in which you need help.

Module	Look here for	eBook page
1.13 Forming and finding	• understanding a claim • classifying elements in the claim • identifying variables, terms and concepts related to the claim • developing a research question • planning the research • refining a research question • putting it all together: sample report	e127
1.14 Analysing	• collecting sufficient and relevant evidence • identifying trends, patterns, relationships in the evidence • assessing limitations of the evidence • putting it all together: sample report	e139
1.15 Interpreting	• assessing if the evidence justifies the claim • presenting an argument whether the claim is justified • reaching a conclusion about the claim • putting it all together: sample report	e146
1.16 Evaluating	• evaluating the quality of the evidence • suggesting any extrapolations of the claim • suggesting any improvements/extensions to the investigation • putting it all together: sample report	e148

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1.1 Biological science

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that science provides a systematic approach for comprehending the world
- recognise that biology is the field of science that investigates living organisms and systems
- understand that biology comprises a range of divisions
- explain the difference between scientific models, theories, laws, principles and facts.

Science is a human endeavour to understand the world and the known universe. It is one method of many used to develop knowledge. This knowledge is continually refined and redefined. Science follows a **methodology** to observe, test, measure, analyse and evaluate natural **phenomena** in a way humans can understand and construct meaning. There is no single **scientific method**; however, it is a cyclical process including observation of natural phenomena, questioning, hypothesising and predicting, experimentation and gathering **data**, refining and altering all previous elements, and theorising (Figure 1.1.1).

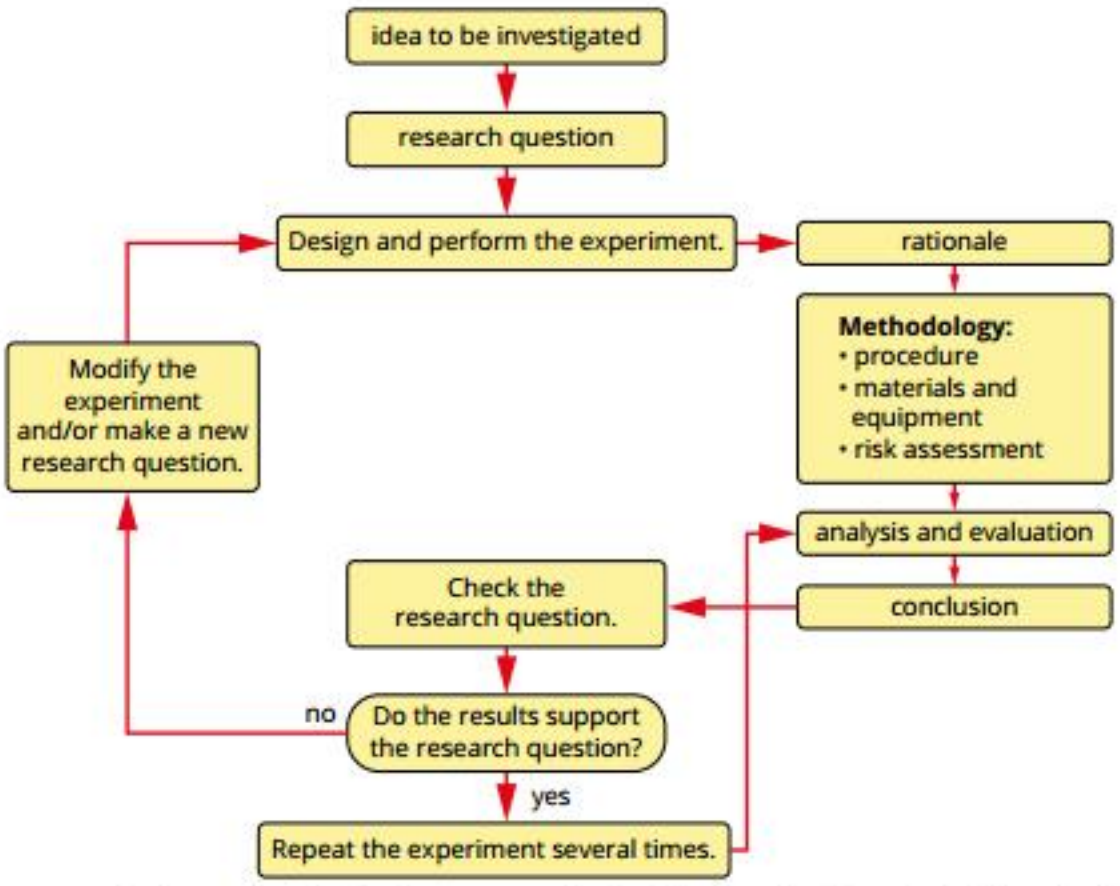


FIGURE 1.1.1 A schematic outlining the processes involved in the scientific method. The scientific method is more of a cyclical process rather than linear.

Biology is the study of living organisms and systems. This includes implies not limited to cells, multicellular organisms and the maintenance and regulation of their internal environments; genetics, reproduction and inheritance; biodiversity and the relationships between the living and non-living; and the continuation of life. Biology can explain how the structure and function of cells interrelate with environmental variables to exchange matter and energy. An understanding of biology informs us about fundamental principles of the living world around us and can inform critical evaluation of social issues and technology.

Today, all scientific disciplines are expanding into new frontiers. The number of divisions and fields in biology is ever-increasing, and the relationship between disciplines is becoming more intertwined. Innovations today are crossing traditional barriers between technology, chemistry, physics and biology of the past 100 or so years.

AREAS OF BIOLOGY

There are numerous areas of biology (Figure 1.1.2), and as technology develops and scientific disciplines become increasingly transdisciplinary, the number of areas increase.

Because living organisms are made up primarily of organic molecules, organic chemistry forms the molecular foundation of the biological sciences and is intimately linked with pharmaceuticals and drug design. Organic chemistry deals with molecules formed predominantly from carbon, hydrogen, oxygen and nitrogen. The range and diversity of organic molecules is enormous—millions of molecules have been identified. Long molecules can be made up of much smaller organic building blocks and form polymers such as the proteins built from 20 different amino acids. Biologically important molecules, such as vitamins, haemoglobin and chlorophyll, also contain inorganic metal atoms.

Traditional divisions in biology are still of vital importance, such as taxonomy (classification of living organisms), and palaeontology (study of fossils). However, much of the exciting current scientific research is interdisciplinary and involves collaboration with many other scientific and technical fields including chemistry, physics, pharmacology, materials science, environmental science and engineering. These types of interdisciplinary collaborations have led to developments in nanotechnology, synthetic biology, bioinformatics, astrobiology, environmental sustainability and many more.

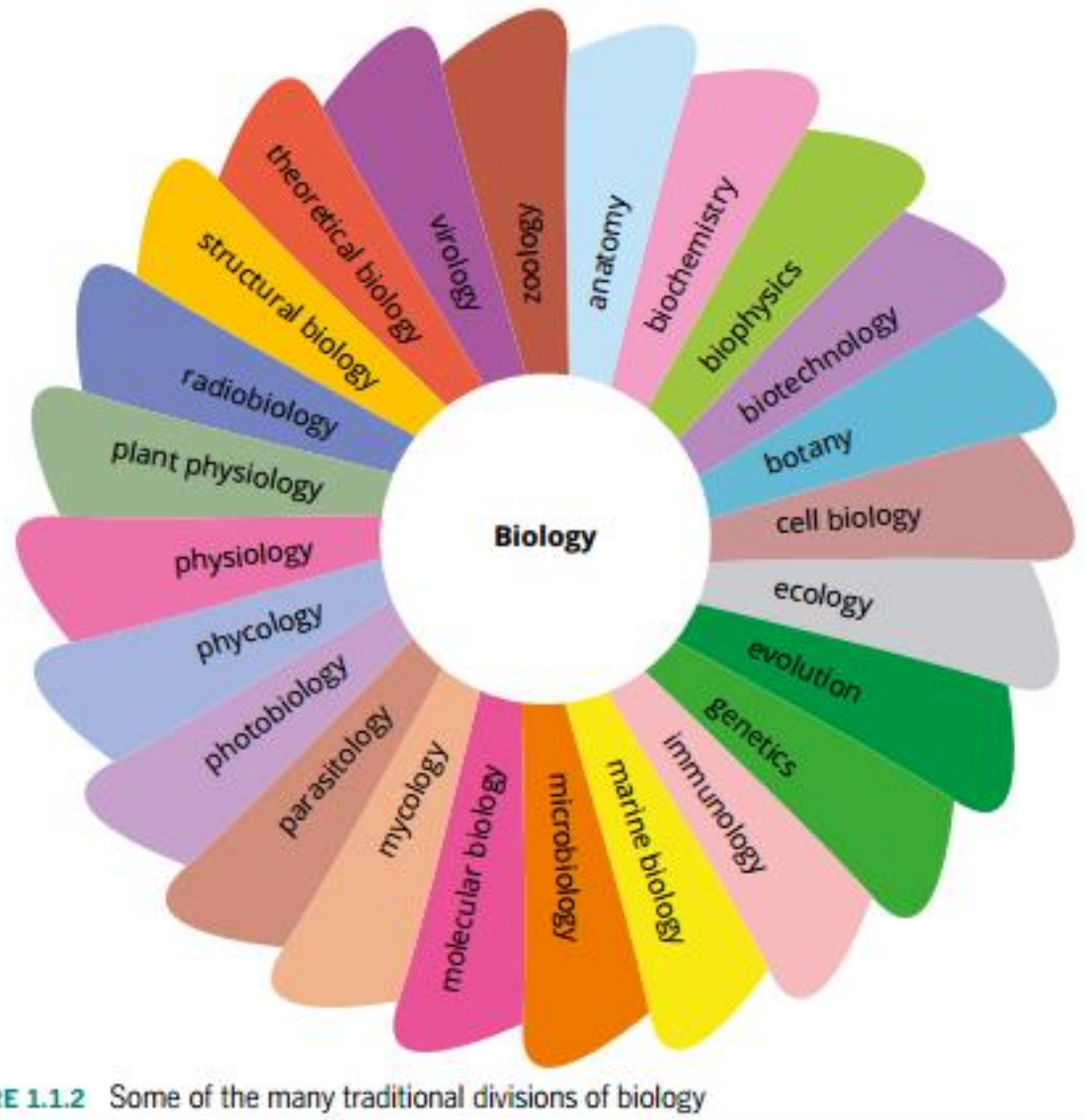


FIGURE 1.1.2 Some of the many traditional divisions of biology

The role and work of a biologist

As the world is changing, science, technology, engineering and maths (STEM) fields are integrating numerous disciplines into vocational roles. A STEM qualification fosters skills in areas such as creativity and critical thinking, and is highly regarded in public and private businesses. The Australian Government's chief scientist report (July 2020) states that 75% of jobs in the fastest growing industries require workers with STEM skills. According to this report, the safest future occupations are STEM related, such as doctors, nurses and teachers. Biological sciences had the equal highest proportion of people working in the public sector across the STEM fields, at 40% (along with Environmental Studies). There are numerous reports every year outlining that the majority of future professions will be STEM related.

These are jobs directly related to a biology qualification and other employment for which biology is a useful background subject (see Table 1.1.1).

The advances in scientific endeavours as well as innovations have seen a significant increase in multiple scientific specialities and disciplines working together. When more than one discipline is required to solve a problem or develop an innovation, it is said to be transdisciplinary. Transdisciplinary approaches to scientific endeavours form the heart of STEM.

TABLE 1.1.1 A degree in biology would be required or beneficial in these jobs

Job	Typical role description
environmental consultant	They work on commercial or government contracts with various environmental issues for clients. This work covers a wide range of disciplines such as assessment of air, land and water contamination, surveys of flora and fauna, environmental impact assessment, environmental audit, waste management, development of environmental policy and management systems.
marine biologist	They specialise in studying the ocean and marine life. They conduct in-depth experiments, do surveys, restore marine environments, rescue and rehabilitate sick or injured marine animals, and monitor marine animal behaviours for research purposes. May involve scuba diving.
medical scientist	Biomedical scientists work alone or in teams in laboratories, conducting research and carrying out tests, operating and developing medical equipment, assisting doctors with diagnoses and developing immunotherapies and vaccines.
sports scientist	They provide expert advice and support to athletes and coaches to help them understand and enhance sports performance, using evidence-based and quality-assured practices to evaluate and develop effective strategies for training, competition and recovery.
forensic scientist	They collect, analyse and provide impartial scientific evidence for use in courts of law to support the prosecution or defence in criminal and civil investigations.
food and nutrition scientist	They cover a wide range of work from scientifically assessing peoples' dietary needs to creating personalised nutrition plans, checking safety standards of food production, developing new food products, laboratory testing for nutritional values, testing of food packaging and storage options.
animal attendant	works directly with animals, providing basic care, exercise and enrichment activities. These attendants perform the daily tasks to keep animals in care appropriately fed and healthy in a veterinary clinic, animal shelter, wildlife park, marine centre and the like.
secondary school teacher or university lecturer	Along with teaching biological sciences to students, lecturers in particular, may also undertake academic or industry research, and may be involved in school or departmental management.
science writer	They research, write and edit scientific news articles and features for environmental, agricultural and professional publications, specialist scientific and technical journals, and the general media.

Some of the careers related to biology can be seen in Figure 1.1.3. The levels shown indicate the amount of training required, with Level 4 being the highest.

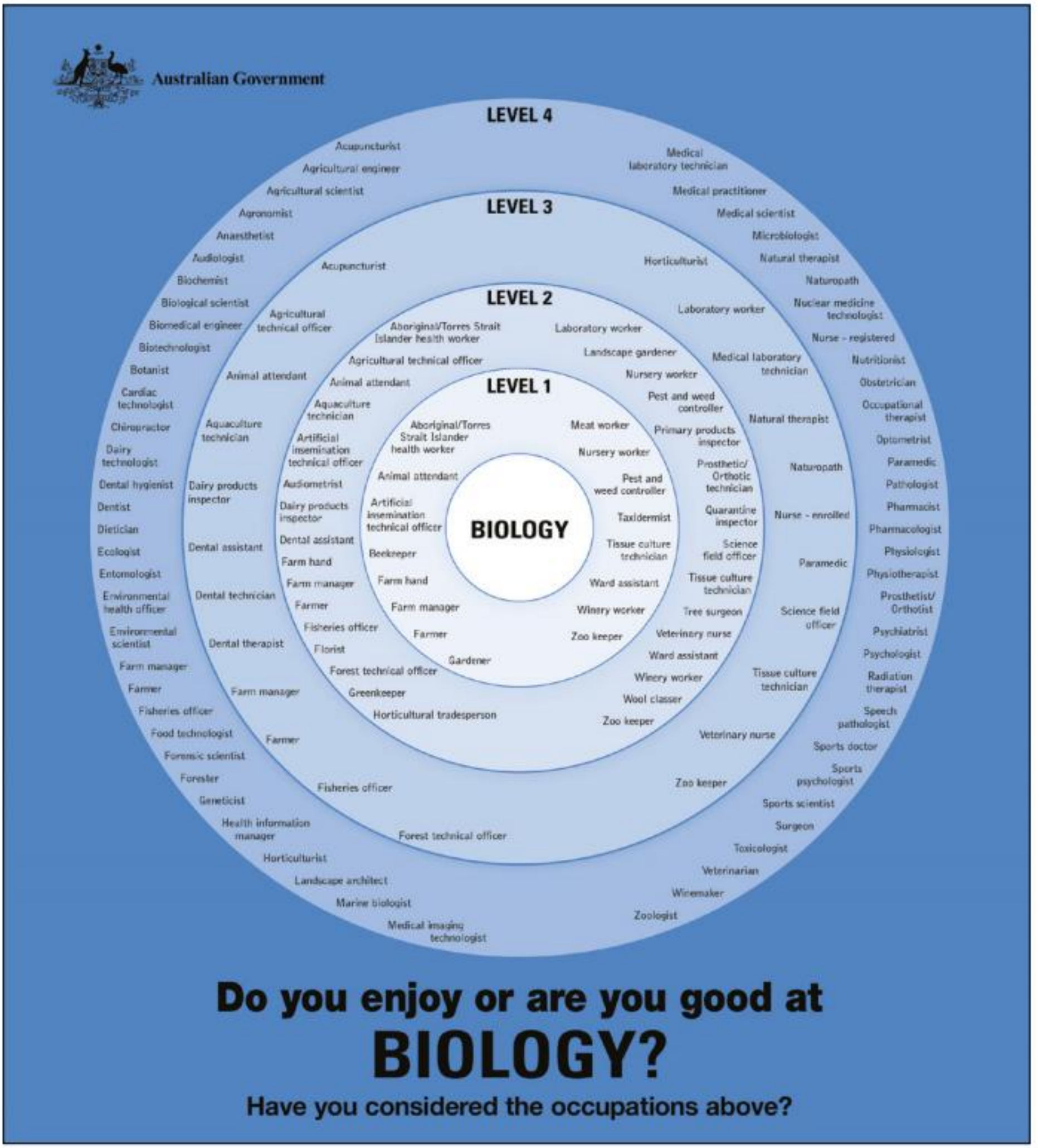


FIGURE 1.1.3 Do you enjoy or are you good at biology? Have you considered these careers?
The diagram shows levels of 1 to 4 which relate to the level of education required for each career.

THE WORK OF SCIENTIFIC RESEARCH

The foundation of all sciences, including biology, is scientific research. Scientific research works in the unknown to develop new understandings. To build evidence, it may take years and many experiments. In interpreting scientific evidence, scientists endeavour to establish models, **theories**, **laws**, **principles** and facts. These are illustrated in Figure 1.1.4.

It is important to note that all these deductions change over time as knowledge develops.

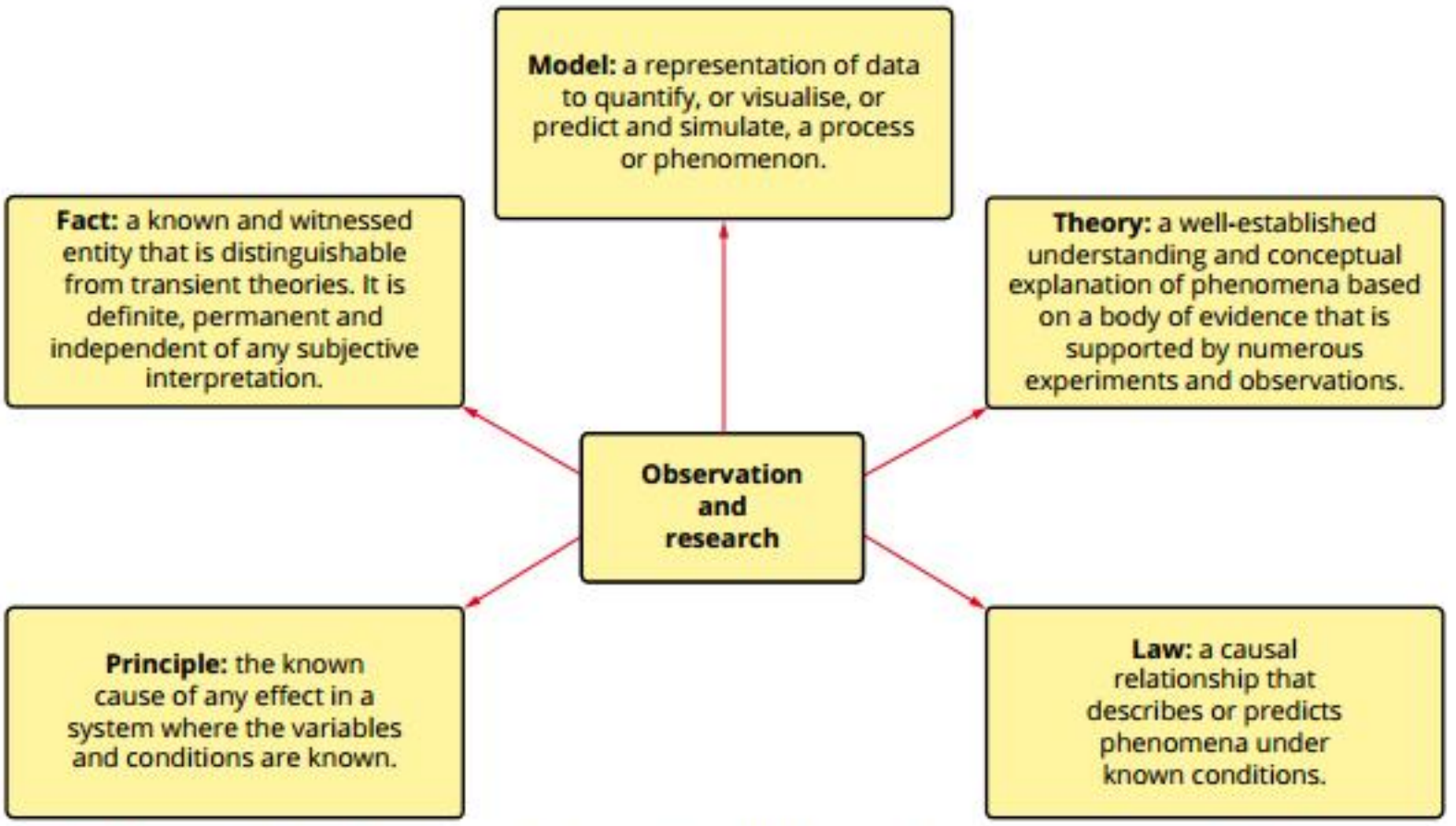

Model: a representation of data to quantify, or visualise, or predict and simulate, a process or phenomenon.
Fact: a known and witnessed entity that is distinguishable from transient theories. It is definite, permanent and independent of any subjective interpretation.
Theory: a well-established understanding and conceptual explanation of phenomena based on a body of evidence that is supported by numerous experiments and observations.
Observation and research
Principle: the known cause of any effect in a system where the variables and conditions are known.
Law: a causal relationship that describes or predicts phenomena under known conditions.

FIGURE 1.1.4 Scientific research and observations are used to establish or develop models, principles, laws, theories and facts.

1.1 Review

SUMMARY

- Science is a systematic approach to develop knowledge through testable explanations and repeatable methods.
- Biology is the study of living organisms and systems.
- There are numerous divisions of biology, and as technology improves and scientific disciplines become increasingly transdisciplinary, the number of divisions grows.
- STEM (science, technology, engineering and mathematics) research is a transdisciplinary approach to scientific endeavours.
- Scientific evidence is used to formulate and establish models, theories, laws, principles and facts.
- A scientific model is an idealised representation of a process, a phenomenon or data.
- A scientific theory is a well-established understanding and conceptual explanation of phenomena based on a body of evidence.
- A scientific law is a causal relationship that describes or predicts phenomena under known conditions.
- A scientific principle is the known cause of any effect in a system where the variables and conditions are known.
- A scientific fact is a known and witnessed entity that is distinguishable from transient theories; facts are definite, permanent and independent of any subjective interpretation.

Part A will consider the basic mathematical skills, data skills and applications that will be required for the student experiment, research investigation, data test and practicals.

1.2 Orders of magnitude and estimation

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- convert numbers to and from scientific notation
- estimate the value of a quantity using its order of magnitude
- estimate the value of a quantity of calculation
- understand that the SI prefixes are multipliers of powers of ten of the base units.

SCIENTIFIC NOTATION

In scientific research and literature, measurements are often written in **scientific notation** to overcome confusion or ambiguity when displaying numbers with magnitude, for example 2.56×10^{-6} (0.000 002 56). It is important to understand the numbers in scientific notation, their order of magnitude and how to convert them into other units.

Quantities are written as a number between one and ten and then multiplied by an appropriate power of ten. Note that 'scientific notation', 'standard notation' and 'standard form' and 'power of ten notation' all have the same meaning.

Examples of some values written in scientific notation are:

- 6.022×10^{23} particles
- $25.25\,\text{mL} = 2.525 \times 10^{-2}\,\text{L}$
- 2.0×10^{-8} m diameter of a virus.

Values given in scientific notation show the significant numbers and indicate the precision of measurement during experimentation by the instrument. Precision and accuracy are covered in more detail in Module 1.5.

SKILLBUILDER 1.2.1

Transforming decimal notation to scientific notation

Scientists use scientific notation to handle very large and very small numbers. For example, instead of writing 0.000 000 035, scientists would write 3.5×10^{-8}.

A number in scientific notation (also called standard form or power of ten notation) is written as:

$a \times 10^n$

where:

a is called the coefficient, and is a real number between 1 and 10, that is $1 \leq a < 10$

n is called the exponent, and is an integer (a positive or negative whole number). n is the power that 10 is raised to.

Note that if n is positive, then the number is larger than one, and if n is negative, then the number is less than 1.

To transform a very large or very small number into scientific notation, perform these steps.

1. Move the decimal place from its position in the original number until you get a number that is greater than or equal to 1 but less than 10.
2. Count the number of places that the decimal point has moved. This number is the exponent – the power of ten (Figure 1.2.1).
   - If the decimal point is moved n places to the left, n will be a positive number; for example $510 = 5.1 \times 10^2$.
   - If the decimal point is moved n places to the right, n will be a negative number; for example $0.051 = 5.1 \times 10^{-2}$.

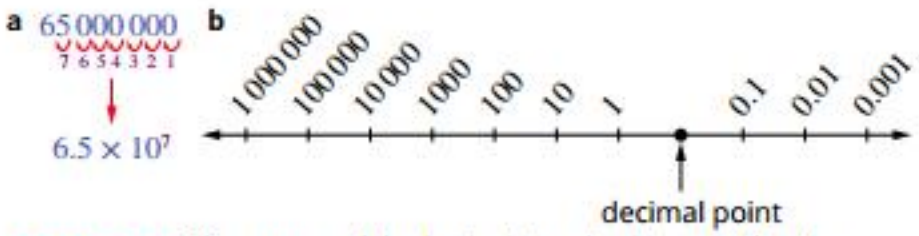


FIGURE 1.2.1 The exponential value is determined by counting the number of places the decimal point needs to be moved.

You will notice from Table 1.2.1 that when large numbers are written in scientific notation, the 10 has a positive exponential value. Very small numbers smaller than 1 are written with 10 to a negative exponent.

Test the skill

Convert 121 000 to scientific notation.

TABLE 1.2.1 Converting from scientific notation

Scientific notation	Number (expanded form)
1×10^{-9}	0.000000001
1×10^{-6}	0.000001
1×10^{-3}	0.001
1×10^{-1}	0.1
1	1
1×10^{1}	10
1×10^{3}	1000
1×10^{6}	1 000 000
1×10^{9}	1 000 000 000

Worked example 1.2.1

CONVERTING TO SCIENTIFIC NOTATION

A typical virus is 0.000 000 020 m in diameter. How many individual viruses of this size can fit across a typical pinhead with a diameter of 1.5 mm?

Thinking	Working
The size of a virus has too many digits for a calculator or will take an excessive time to calculate, so convert it to scientific notation.	
Move the decimal place to the final digit.	0.000 000 020 = 2.0
Count the number of places the decimal placed was moved.	8
Use the number of decimal places moved as the power exponent to base 10.	2.0×10^{8}
Determine whether the decimal was moved left or right.	right
Place a negative symbol (–) in front of the power of 10.	2.0×10^{-8} m
Convert 1.5 mm to m.	1.5 mm = 0.15 cm = 0.0015 m
Convert to scientific notation, using the same steps as above.	1.5×10^{-3} m
Calculate the number of individual viruses that can fit on a pinhead by dividing the pinhead diameter (1.5×10^{-3}) by the virus diameter (2.0×10^{-8}).	$\text{number} = \frac{1.5 \times 10^{-3}}{2.0 \times 10^{-8}}$ = 75 000

➤ Try yourself 1.2.1

CONVERTING TO SCIENTIFIC NOTATION

Escherichia coli is a common disease-causing bacterium. It averages approximately 0.000 002 m in length. Calculate how many *E. coli* bacteria could fit across a pinhead if they were lined up length to length.

You should be routinely using scientific notation to express very large and very small numbers. This also involves learning to use your calculator intelligently. Scientific and graphics calculators can be put into a mode whereby all numbers are displayed in scientific notation. It is useful when doing calculations to use this mode rather than frequently attempting to convert to scientific notation by counting digits on the calculator display.

An important reason for using scientific notation is that it removes ambiguity about the precision of some measurements. For example, a measurement recorded as 240 g could be a measurement to the nearest gram; that is, somewhere between 239.5 g and 240.5 g, or it could have been rounded up to the nearest 10 g. By writing this quantity as 2.40×10^2 g, it makes clear the accuracy of the value (in this case, 3 **significant figures**).

Writing numbers from scientific notation

To write a number into long form from scientific notation, follow these rules.

- If the exponent is positive, write the coefficient without the decimal point. Continue to write zeros after the last digit in the coefficient so that the number of placeholders after the first digit is equal to the exponent.
- If the exponent is negative, write a zero in the units placeholder, then a decimal point. Continue to write zeros until the total number of zeros (including the zero in the units placeholder) is equal to the value of the exponent. Then write the coefficient without the decimal place.

Worked example 1.2.2

WRITING QUANTITIES FROM SCIENTIFIC NOTATION

The mass of one vaccinia virus is calculated as 1.675×10^{-17} kg. Write this value in long form.

Thinking	Working
The exponent is negative, which means the number in long form will be less than 1. Write a zero in the units column and then a decimal point. After the decimal point, write another 16 zeros followed by the coefficient without the decimal place.	0.000 000 000 000 000 016 75 kg

➤ Try yourself 1.2.2

WRITING QUANTITIES FROM SCIENTIFIC NOTATION

Write the following values in long form:

a the diameter of a magnesium ion is 144×10^{-12} m

b the number of hydrogen molecules in 1 mole of hydrogen gas is 6.02×10^{23}.

Scientific notation is the preferred way of communicating very large and very small measurements and information in biology and biochemistry. Biologists measure the number of DNA bases in a genome in the millions or the size of biological molecules that are a millionth or a billionth of a metre. It is important to understand the numbers in scientific text and their order of magnitude and be able to convert them to other units. For convenience and ease of use, and to avoid mistakes with numerous zeros, quantities in science problems should first be converted into scientific notation before any calculations can be carried out.

ORDERS OF MAGNITUDE, PREFIXES AND UNITS

Two methods are used to write down a physical measurement: prefixes and scientific notation.

The other form of communicating measured results is to use units that represent the order of magnitude. If 2.56×10^{-6} m was measured, it could be written either in the scientific notation as it is already, or as 256 μm. The symbol μ represents the prefix *micro* (Table 1.2.2), which means 'one millionth of the measured unit'. So, when *micro* is placed in front of metre (as a prefix), it becomes micrometre, meaning 'one millionth of a metre', therefore 256 μm becomes 256 millionths of a metre. Again, these are very small measurements and numbers.

An International System of units (SI) **prefix** is a symbol written at the front of a unit to indicate that the measurement is a power of ten higher or lower than the unit itself.

There are twenty SI prefixes in use and most of them denote units that are one thousand (10^3) times larger or smaller than the previous prefix. Table 1.2.2 lists the twelve SI prefixes commonly used in biology. You should become familiar with both scientific notation and prefixes for units of measurement. You should also be able to interchange them.

TABLE 1.2.1 The 12 official SI prefixes used in biology

Prefix	Symbol	Multiplier	Description	Decimal	Example
tera	T	10^{12}	trillion	1 000 000 000 000	1 TByte hard drives are now common.
giga	G	10^9	billion	1 000 000 000	Over 10 Gigatonnes of carbon dioxide is produced from burning coal.
mega	M	10^6	million	1 000 000	1 MHz (megahertz) is close to the frequencies at which TV stations broadcast.
kilo	k	10^3	thousand	1000	Used for body weight or buying food e.g. 5 kg of potatoes. 1 kg is 1000 g.
hecto	h	10^2	hundred	100	1 hPa (hectopascals) meteorologic unit for atmospheric pressure: typical sea-level air pressure is ~1013 hPa.
deca	da	10^1	ten	10	1 daN (decanewtons) is approximately the force exerted by a 1 kg object on the surface of the Earth.
–	–	1	unit	1	The base unit e.g. metre or gram.
deci	d	10^{-1}	tenth	0.1	1 dL a common unit of measurement in blood screening for health conditions. 1 dm^3 = 1 L.
centi	c	10^{-2}	hundredth	0.01	30 cm pencil case ruler. 1 cm equals 1/100th (0.01) of a metre.
milli	m	10^{-3}	thousandth	0.001	1000 mm in 1 metric ruler and 1000 mL in 1 Litre of milk
micro	μ	10^{-6}	millionth	0.000 001	μg is commonly used as the unit for medicine dosage.
nano	n	10^{-9}	billionth	0.000 000 001	The wavelength of yellow sodium streetlights is 550 nm.
pico	p	10^{-12}	trillionth	0.000 000 000 001	Blood concentrations of insulin are measured in picomoles (pmol).

SI prefixes are used to avoid writing very small or very large numbers with lots of zeros. The prefix is added directly to the name of a unit, and a prefix symbol attaches directly to the symbol for a unit.

Leave a space between the last number and the unit but no space between the prefix and unit symbols: 30 cm is correct. It is not correct to write 30cm or 30 c m. The larger prefixes are written as a capital letter, while the smaller prefixes are written using a lowercase letter. It is important to give a symbol the correct case (upper- or lowercase). There is a big difference between 1 mm and 1 Mm.

Note that the fundamental unit kilogram contains a prefix, but all multiples of mass are written with the base unit grams. As an example, one million kilograms is not written as one megakilogram, but as one gigagram (Gg).

Before any measurements with a prefix can be substituted into a mathematical formula, they must be converted into SI units without the prefix, that is, written in scientific notation.

When converting units, it is useful to write the conversions as fractions and then cross-cancel out to ensure that the conversion is done correctly.

SKILLBUILDER 1.2.2

Converting between units

Often in science you need to convert from one unit to another to complete a calculation. Knowing how to change between units is an important skill; however, conversion factors should be used carefully. As some units of measurement are difficult to visualise, knowing the size of different units in relation to each other will aid your understanding and help you avoid errors in your calculations. The most common mistake made with conversion factors is multiplying rather than dividing.

Cortisol is a natural hormone found in the body; however, it is used by some athletes to increase performance. Normal levels of cortisol in the blood are around 400 nmol L^{-1}. To put this into context, you can convert this value into a unit you can easily visualise, like grams per litre (g L^{-1}).

To convert 400 nmol L^{-1} to mol L^{-1} (before converting to g L^{-1}), you need to move the decimal seven places to the left, so it becomes 0.000 000 4 mol L^{-1}.

This can also be written as 4.0×10^{-7} mol L^{-1}.

One mole of cortisol weighs 362.46 g, therefore 0.000 000 4 mol L^{-1} is equal to 0.000 145 g L^{-1} (or 1.45×10^{-4} g L^{-1})—very small indeed!

A mole (symbol: mol) is the SI unit to quantify a substance. For a biology student, it is sufficient to just follow the calculation, not to understand the concept (frequently used in chemical science and defined as the amount of a substance that contains as many particles as there are atoms in exactly 12 g of carbon-12 isotope, also known as Avogadro's number, 6.022×10^{23} particles).

Worked example 1.2.3

CONVERTING BETWEEN UNITS

Convert 60 700 grams to kilograms.	
Thinking	**Working**
Convert the units from grams to kilograms.	$= \frac{g}{} \times \frac{kg}{g}$ $= \frac{\cancel{g}}{} \times \frac{kg}{\cancel{g}}$ $= kg$
To convert grams to kilograms, multiply 60 700 by kg per g. Since 1 kg = 1000 g, the kg is replaced by 1 and the g is replaced by 1000.	$= \frac{60\,700}{} \times \frac{kg}{g}$ $= \frac{60\,700}{} \times \frac{1}{1000}$ $= 60.7\ kg$

➤ Try yourself 1.2.3

CONVERTING BETWEEN UNITS

The photosynthetic rate of CO_2 by a plant was measured to be 6.2 $\mu mol\, m^{-2} s^{-1}$. Calculate the number of moles of CO_2 that are photosynthesised per square metre per second.

When converting from one unit to another it is essential that you check that the answer makes sense. For instance, if converting from mm to km, you would expect that there are many mm (millimetres) in a km (kilometre). The inversion of prefix conversions is one of the major sources of mistakes in general science calculations.

1.2 Review

SUMMARY

- 'Scientific notation', 'standard notation', 'power of ten notation' and 'standard form' all have the same meaning.
- A number in scientific notation is written as $a \times 10^n$, where:
  - a is called the coefficient, and is a real number between 1 and 10; that is, $1 \leq a < 10$
  - n is called the exponent, and is an integer (positive or negative whole number). n is the power that 10 is raised to and is sometimes called the index value.
- A range of scientific prefixes can alternatively be used to represent 10^n. Some examples are milli-, micro-, kilo-.

KEY QUESTIONS

Describe

1 State the prefix and symbol for the following orders of magnitude.
- **a** 1000000000
- **b** 0.000001
- **c** 0.000000001

2 Identify what the following units mean.
- **a** μ
- **b** p
- **c** k

3 Explain why scientists communicate using scientific notation.

Apply

4 Convert the following to scientific notation.
- **a** 0.000000000012
- **b** 0.00045
- **c** 73000000000000000

5 Represent the following values in scientific notation.
- **a** 637 nm
- **b** 319 s

6 Write the expanded form of the following numbers.
- **a** 1.1×10^{-10} m
- **b** $7.031 \times 10^{6}\, m\, s^{-2}$

7 Determine the order of magnitude of the following values.
- **a** 6.41×10^{9} m
- **b** $1.201 \times 10^{-3}\, m^3$

8 Match each number with its scientific notation.

Number	Scientific notation
0.002	2×10^{3}
2000	1.234×10^{-1}
0.1234	2×10^{-3}
123.4	1.234×10^{1}
12.34	1.234×10^{2}

Analyse

9 Convert the following lengths into the unit in brackets, presenting the information in scientific notation.
- **a** 1.56 mm (μm)
- **b** 8.35 nm (cm)

10 A human hair is about 50 μm wide, whereas a virus is 2×10^{-8} m wide. How many times wider than the width of the virus is the width of the human hair?

1.3 Mathematical basics for biology

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that the rules of mathematics apply for solving mathematical operations in biology
- perform basic mathematical operations using percentages and fractions
- rearrange and substitute formulas using algebraic rules
- use the equation of a straight line to determine its gradient and *y*-intercept.

Science uses mathematics in working with data. The evidence can be quantified, measured and analysed. This is known as statistical analysis (a branch of mathematics).

When conducting mathematical operations or calculations, remember all the rules of mathematics apply, including:

- order of operations
- rules for transposing formulas and working with fractions
- substitutions
- approximations.

EXAMPLES OF MATHEMATICAL BASICS IN BIOLOGY

Measuring cells under a compound light microscope

A compound light microscope can be used to examine in detail thin sections of plant and animal tissues and determine the size of the cell and its visible components (Figure 1.3.1 e21). To do this, you need to determine the magnification and field of view of the microscope.

Total magnification of a microscope

The total magnification of the microscope is calculated by multiplying the magnifying powers of the objective and the eyepiece lenses. The eyepiece (or ocular lens) of a microscope is the lens closest to the eye, and usually magnifies objects by 10 times (10×) their actual size. The other lens is the objective lens and is located on the rotating part of the microscope barrel closest to the stage and specimen. There are usually three or four objective lenses, each allowing for a different degree of magnification. The magnification value is marked on the side of each lens.

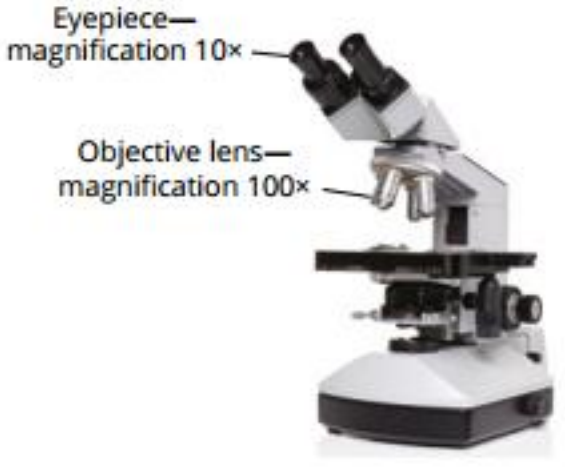
For example, a 10× objective used with a 10× eyepiece gives a total magnification of 100×.

Worked example 1.3.1 demonstrates examples of the use of mathematics in biology.

Worked example 1.3.1

MATHEMATICAL BASICS IN BIOLOGY

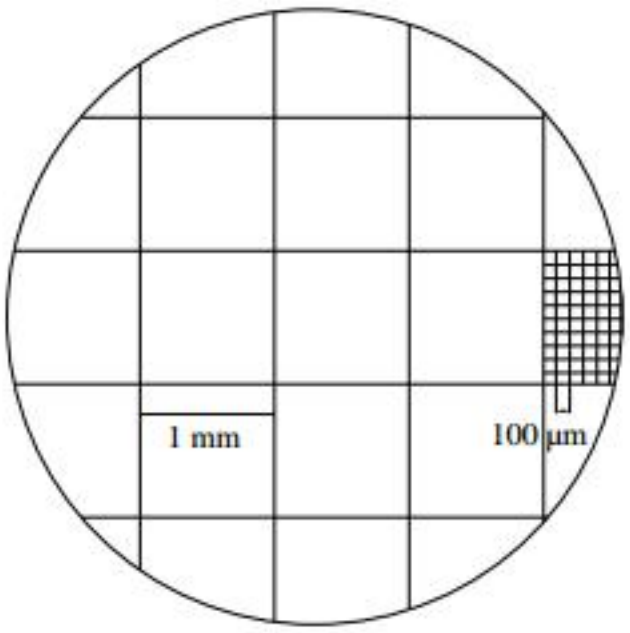
Calculating the magnification of a microscope



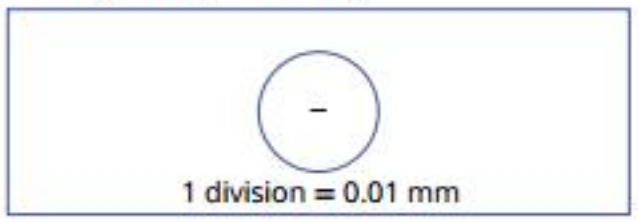
Thinking	Working
Record the magnification of each lens: ocular (eyepiece) and objective. Write down the calculation. Substitute in the values.	$M_t = M_e \times M_{ol}$ $= 10^1 \times 10^2$ $= 10^{1+2}$ $= 10^3$ or 1000x M_t = total magnification M_e = eyepiece magnification M_{ol} = objective lens magnification

To estimate the size of specimens viewed, you need to calculate the field of view under the microscope (the circle of light visible when looking through the ocular lens). Also, all biological drawings require a scale. You can measure the initial field of view by using a stage micrometer slide, which is a mini-grid on a microscope slide.

Calculating the field of view diameter and converting from magnification 40× to magnification 400×



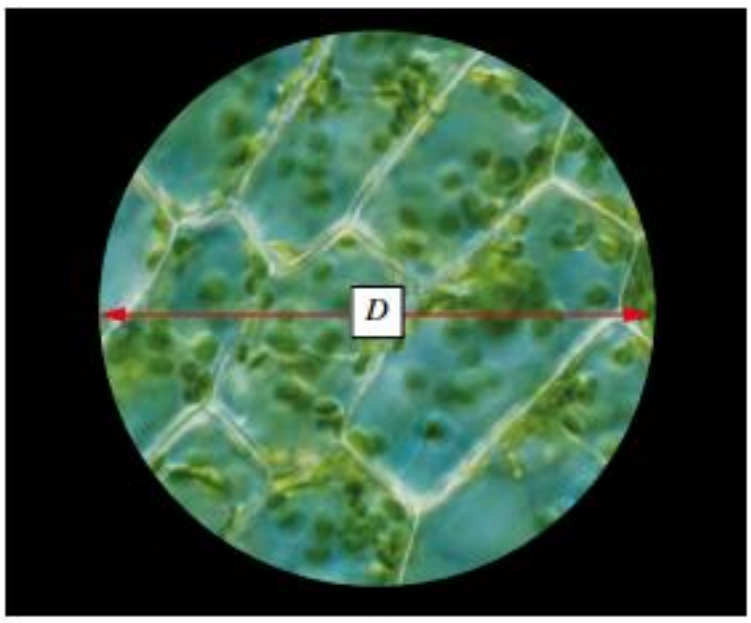
The field of view of this microscope using the 40x lens is 4.6 mm (or 4600 µm). You can work this out by using the mini-grid.



100 × 0.01 mm = 1 mm

Thinking	Working
To determine the size of microbes or specimens under the microscope, you need to know the field of view diameter. The diameter for this field of view at magnification of 40× is 4.6 mm.	$D_b = \frac{M_a}{M_b} \times D_a$ D_a = original diameter D_b = new diameter M_a = original magnification M_b = new magnification To calculate the field of view diameter at 400×: $D_{\times 400} = \frac{M_{\times 40}}{M_{\times 400}} \times D_{\times 40}$ $= \frac{40}{400} \times 4.6$ $= 0.46$ mm

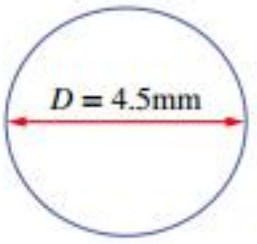
Calculating the size of a specimen in the microscopic field of view



× 400

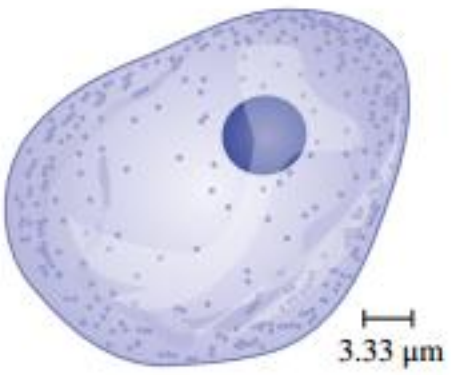
Thinking	Working
To complete a comparative study, estimate organelle size, determine organelle density (or concentration) and sometimes calculate rates of cellular processes for cells, you need to know the size of a specimen. Determine the field of view. Decide if width or length of each cell is to be counted. Count the specimens that fit across.	If the field of view diameter at 400× is 0.4 mm and using length, 2.5 cells fit across the diameter. Size (length) = $\frac{D}{\text{no. of speciment across } D}$ $= \frac{0.4 \text{ mm}}{3}$ $= 0.13$ mm

Calculating the area of a microscopic field of view



Thinking	Working
For bacterial studies, including lawn cultures and colonies, it is difficult to count the number of bacteria so you use the area of population. Use the same calculation as for area of a circle ($A = \pi r^2$) Convert diameter (D) to radius (r).	$A = \pi r^2$ $= \pi 2.25^2$ $= 15.896\,25\ mm^2$ $= 15.9\ mm^2$ (considering significant figures, see Module 1.5) r = the radius (half of D the diameter) π = the mathematical constant pi, a key on your calculator, or use 3.14 or 22/7

Calculating cell size using a scale bar

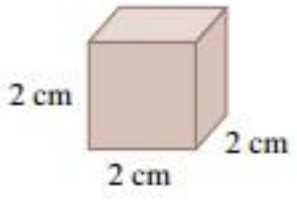


Human cheek cell, unknown magnification

Calculate the length of the human cheek cell to the left.

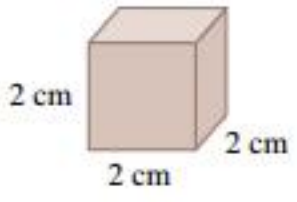
Thinking	Working
Measure the scale bar.	0.5 cm
Convert to µm.	Every 0.5 cm = 3.33 µm
Measure the length of the cell image using a ruler.	The cell is approximately 4 cm in length. $\frac{4}{0.5} = 8, 8 \times 3.33 = 26.64,$ The cell is approximately 27µm across
Convert the length of the cell to µm.	4 x 2 x 3.33 = 26.64 µm

Calculating the surface area (SA) of a cube



Thinking	Working
Dimensions of cells (or simulated cells) are required when determining a cell's capacity to undergo diffusion. SA = (length x length) x 6 sides	SA = (length x length) x 6 sides = (2 cm x 2 cm) x 6 = $24\ cm^2$

Calculating the surface area to volume ratio (SA:V) of a cube



Thinking	Working
This is required to understand the relationship between cell size and rate of diffusion. This also helps to understand the structure and function relationship for cells. SA:V = (length x length) x 6 sides:length^3	SA:V = (length x length) x 6 sides:length^3 = $2^2 \times 6 : 2^3$ = 24:8 divide by 8 (highest common factor) = 3:1 (in simplest form)

Increase field of view = decrease magnification
Decrease field of view = increase magnification

➤ Try yourself 1.3.1a

CALCULATING TOTAL MAGNIFICATION OF A MICROSCOPE

Calculate the total magnification of a light microscope if a 10× ocular lens and a 40× objective lens were used.

➤ Try yourself 1.3.1b

MATHEMATICAL BASICS IN BIOLOGY

A cubic shaped cell has 6 equal sides of 5 mm length × 5 mm width. Calculate:

a the surface area of the cell

b the SA:V ratio of the cell

Objective lens	Total magnification with 10x eyepiece	Field of view
4×	40×	4.5 mm
10×	100×	1.8 mm
40×	400×	450 µm
100×	1000×	180 µm

School microscopes typically have the magnifications and fields of view shown in the table and Figure 1.3.1. Microscopes usually have 10× eyepieces. The total magnification is the product of the eyepiece and objective lenses. The field of view units will differ for lower and higher magnifications. Measurements will typically be in millimetre (mm) or micrometres (µm, 10^{-6}, one millionth of a metre and one thousandth of a millimetre).

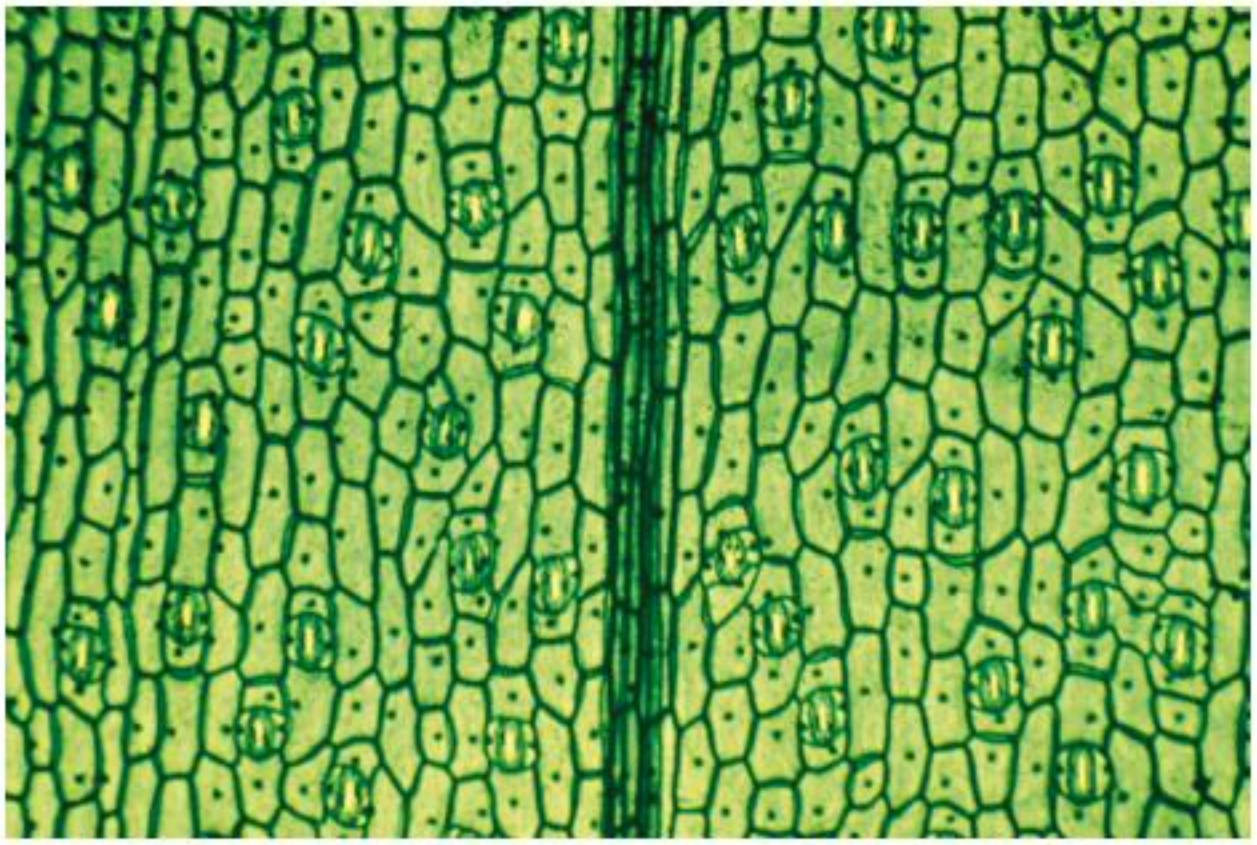


FIGURE 1.3.1 Leaf epidermal cells under a high power objective lens (40×) with total magnification 400×. The field of view is 450 µm. There are about 30 cells across the field of view, so the average width of these cells is about 450/30 = 15 µm.

ANALYSING STRAIGHT LINES

A straight line is the simplest relationship between two variables. The **dependent variable** is plotted on the *y*-axis and the **independent variable** is on the *x*-axis then the relationship between the two variables is represented by the gradient. The *y*-intercept and gradient can be easily calculated by hand or with technology. Figure 1.3.2 shows a typical straight line relationship.

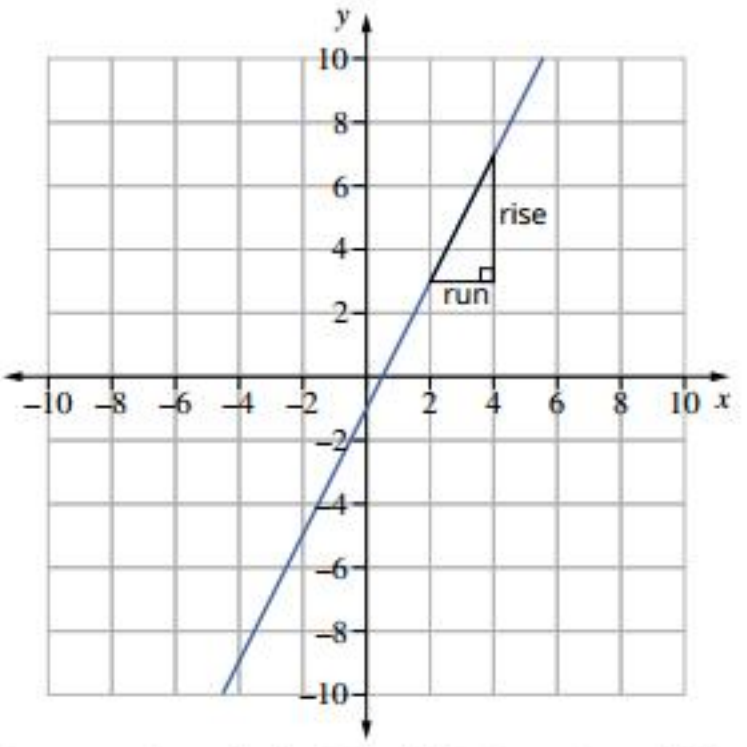


FIGURE 1.3.2 A graph representing a simple relationship between two variables

The gradient of any straight line is calculated using:

$$\text{gradient, } m = \frac{\text{rise}}{\text{run}} = \frac{\Delta y}{\Delta x}$$

The y-intercept, c, is read off the y-axis.

The equation of any straight line is determined by the gradient and y-intercept:

$$y = mx + c$$

In the example in Figure 1.3.2, the equation of the straight line is $y = 2x - 1$ with a gradient of 2 (the rise is $7 - 3 = 4$ divided by the run $= 2$) and y-intercept of -1.

SKILLBUILDER 1.3.1

Interpreting the slope of a linear graph

Scientists often represent a relationship between two variables as a graph. For directly proportional relationships, the variables are related to each other by a straight line, where the slope (or gradient) of the line represents the constant of proportionality between the two variables.

The slope or gradient of the line is defined as the ratio of change between two points in the vertical axis (Δy), divided by the change between two points in the horizontal axis (Δx). In other words, it measures the rate at which one variable (the dependent variable) changes with respect to the other (the independent variable).

The graph below has two straight lines with different slopes (Figure 1.3.3). The steeper slope (solid line) indicates that the rate of change is higher. This means the change is happening more quickly. On the other hand, the flatter slope (dashed line) indicates that the rate of change is lower. This means the change is happening more slowly.

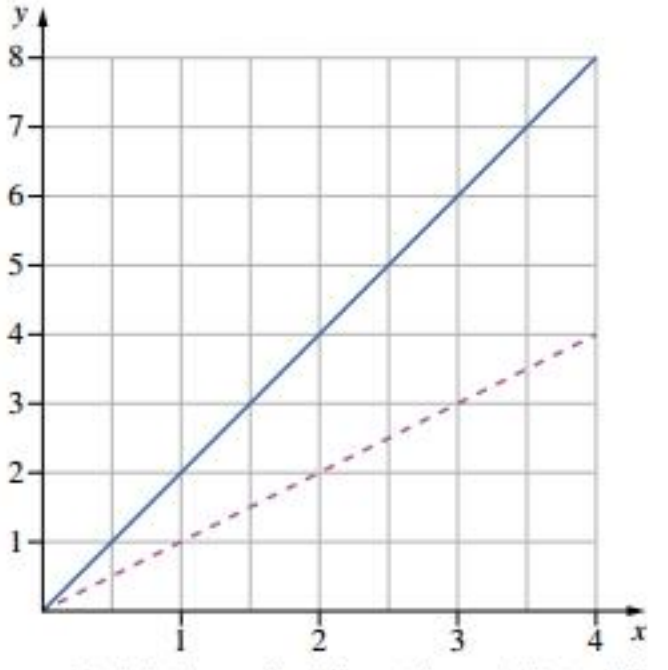


FIGURE 1.3.3 Straight lines with different slopes indicate different rates of change between points on a graph.

Worked example 1.3.2

INTERPRETING THE SLOPE OF A LINEAR GRAPH

On a graph with two sloped lines, identify what the steeper sloped line indicates.

A a faster rate of change

B a slower rate of change

C the same rate of change

D a much slower rate of change

Thinking	Working
The slope or gradient of the line is defined as the ratio of change between two points in the vertical axis (Δy), divided by the change between two points in the horizontal axis (Δx).	A is correct. The steepness of the slope indicates the rate of change. A line with a steeper slope indicates a faster rate of change.

➤ Try yourself 1.3.2

INTERPRETING THE SLOPE OF A LINEAR GRAPH

Identify which of the following options indicates the rate of change of a straight line on a graph:

A area under the graph

B *y*-intercept

C *x*-intercept

D gradient

1.3 Review

SUMMARY

- When conducting mathematical operations or calculations, remember all the rules of mathematics apply, including:
  - order of operations
  - rules for transposing formulas and working with fractions
  - substitutions
  - approximations.
- Mathematics is frequently used in biology for calculations with microscopy and graphing work.

KEY QUESTIONS

Describe

1 Name three ways that mathematics can assist in the study of biology.

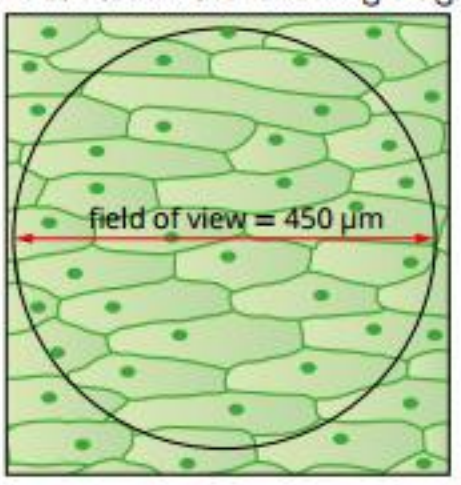
Apply

2 **a** Calculate the missing figures to complete the following table.

Ocular lens	Objective lens	Total magnification
×10	×4	
	×10	×100
×10		×400
×10	×100	

b Which magnification and field of view would be best for viewing cells about:
- **i** 20 µm long
- **ii** 0.7 mm in size?

3 A microscope was set up to view some cells as illustrated in the following diagram.



a The microscope is set up with an ocular lens of ×10 magnification and an objective lens of ×4 magnification. Calculate the total magnification.

b Looking down the microscope, the field of view is determined to be 450 µm. Calculate the actual length of each cell measured in µm.

Analyse

4 Calculate the field of view area of a microscope, for the following field of view diameters.
- **a** 1.4 mm
- **b** 445 µm

5 Calculate the surface area to volume ratio of a block measuring 5 µm × 4 µm × 8 µm.

6 A student recorded the following temperatures of water as it was heated.

Plot a simple straight line graph from the data.

Time (min)	Temperature (°C)
0	22
2	34
4	43
5	50
8	64
10	78

a Calculate the average temperature change per minute during the experiment.

b Determine whether the increase was constant throughout the experiment.

7 The following diagrams represent the field of view visible when various cell types are viewed at different magnifications. The lines in the grids are 100 µm apart.

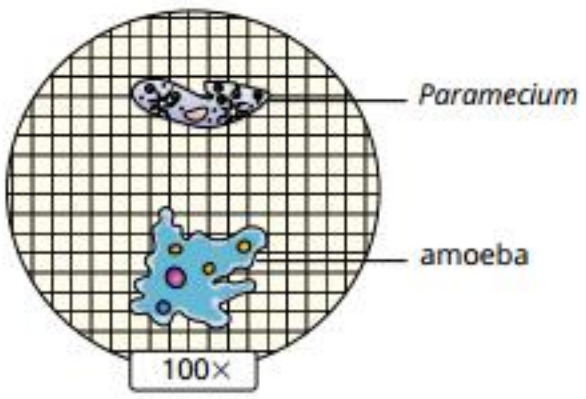
The names of the cell types in the diagrams A to F are in the list below:

phloem, palisade, *Diplococcus*, epidermal, *Spirilli*, cheek cells, human skin cells, *Streptococcus*, red blood cells, amoeba, *Paramecium*, *Bacillus*

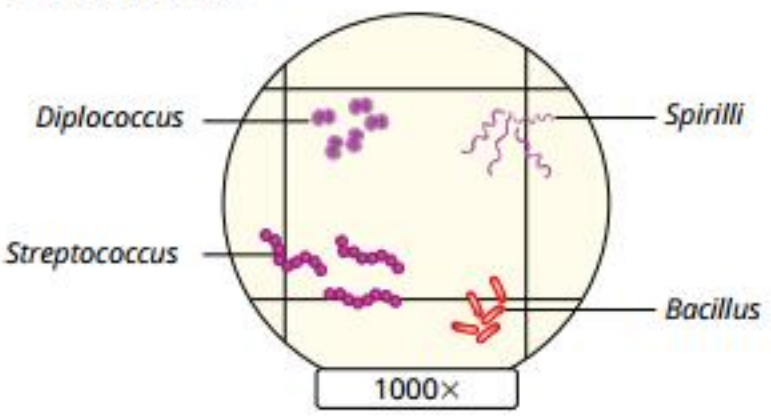
a Use the information in each image to estimate the length of each type of cell from the below. In this instance, the length of the cell is the longest dimension of the cell, top to bottom or left to right.

b Organise the cells in the list in order of size from smallest to largest.

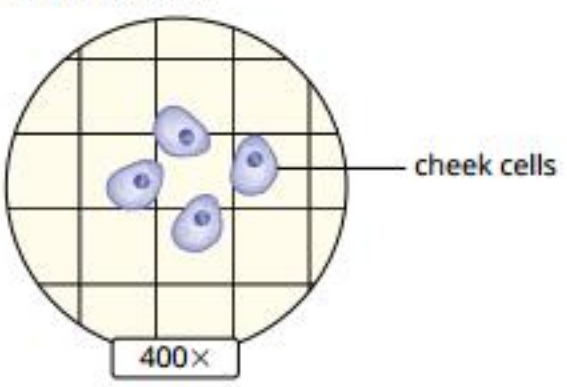
A Protozoans



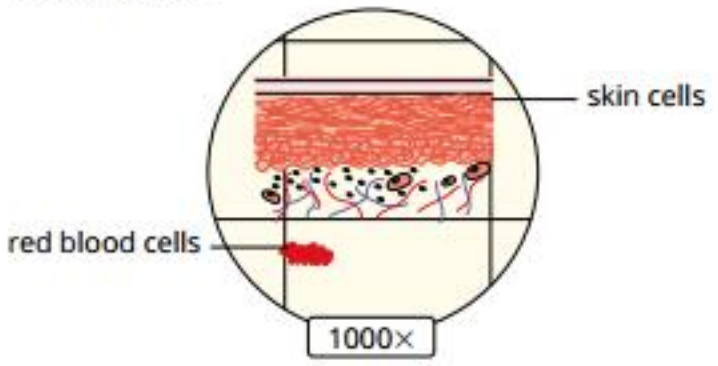
B Bacterial cells



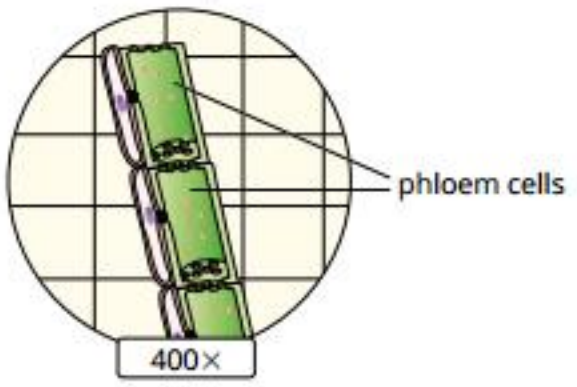
C Animal cells



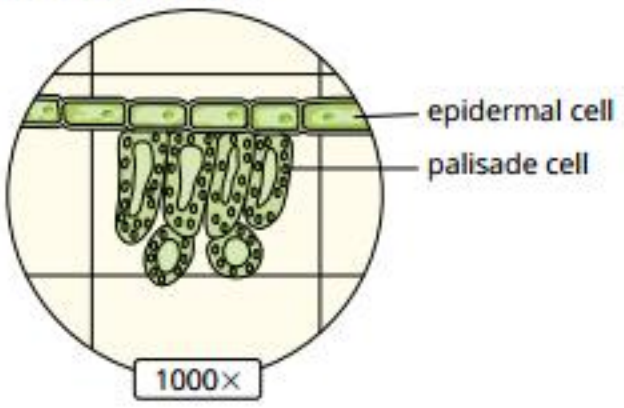
D Animal cells



E Cells from a plant stem



F Cells from the leaf of a plant



1.4 Units

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- appreciate that biology uses the International System (SI) of units
- recall fundamental units, their symbols and definitions
- write measurements and communicate quantities using SI units
- convert between various units of the same type using SI and some non-SI units.

Science uses a specific system of units to describe measurements that are made. In all sciences, measurements are made using the **International System (SI)** of units (from the French name: Système International d'unités). Most countries around the world use and understand these units as they are internationally accepted in science, industry, construction, commerce, sport and everyday life.

MEASUREMENT AND UNITS

Make sure the units you measure or observe are accurate and remember to use SI units when recording your experimental evidence. Table 1.4.1 presents common quantities and SI units used in biology. Three temperature scales are commonly used: the SI unit is Kelvin (K), however the unit Celsius (°C) is routinely used in Australia. (Figure 1.4.1).

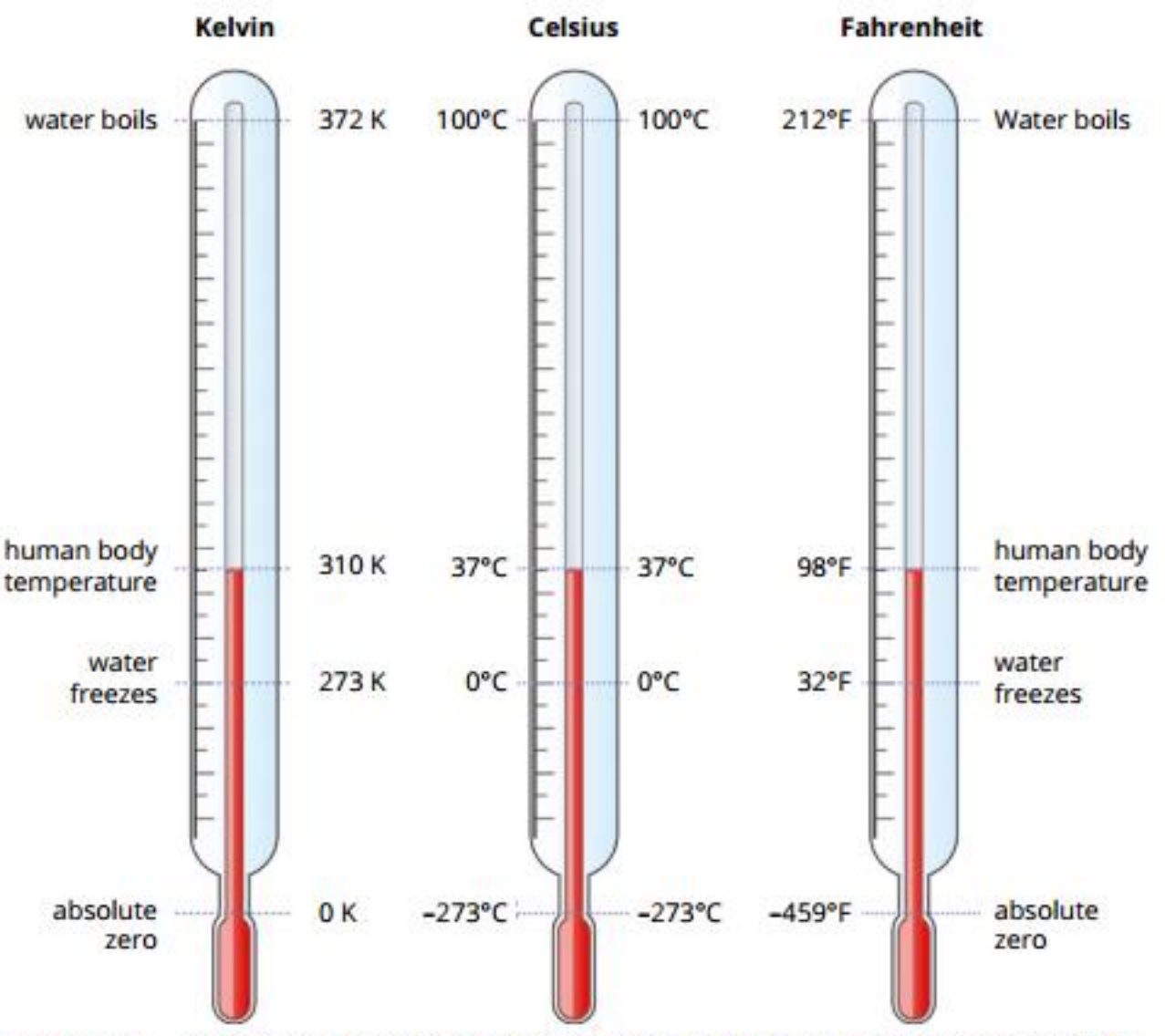


FIGURE 1.4.1 Comparison of the three temperature scales: Kelvin, Celsius, Fahrenheit. Celsius is the scale used for school experiments and for everyday life in Australia

TABLE 1.4.1 Common quantities and SI units used in biology

Quantity	SI symbol for quantity	SI unit	Other commonly used unit (symbol)
mass	*m*	gram (g) kilogram (kg)	
volume	*V*	litre (L) millilitre (mL)	
energy	*E*	joule (J) kilojoule (kJ)	calorie (cal) used in food nutrition 1 kilocalorie equals 4.184 kilojoules
concentration (molarity)	*c*	moles per litre (mol L^{-1}, M or mol dm^{-3})	grams per 100 mL (g/100 mL))
per cent solution	%	weight per volume (% w/v) weight per weight (% w/w) volume per volume (% v/v)	
density	*d*	g mL^{-1} or kg L^{-1}	
temperature	*T*	Celsius (°C), Kelvin (K)	Fahrenheit (°F) still used in USA
pressure	*P*	atmospheres (atm), pascals (Pa), millibar (mb)	millibars are used in meteorology. Average air pressure at sea level of 1 atm = 1013.2 mb.
time	*t*	second (s)	minute (min) hour (h/hr)
distance (length)	*d*	metre (m)	millimetre (mm), kilometre (km)

Correct use of unit symbols

The correct use of unit symbols removes ambiguity, as symbols are recognised internationally. Some important points to keep in mind when writing units and recording measurements and numbers are given in Table 1.4.2.

Uppercase letters are only used for the *symbols* of the units that are named after people. For example, the unit of energy is joule and the symbol is J. The joule was named after James Joule who was famous for studies into energy conversions. The exception to this rule is 'L' for litre. Uppercase 'L' is used because a lowercase 'l' looks like the numeral '1'.

The product of a number of units is shown by separating the symbol for each unit with a space. The division or ratio of two or more units can be shown in fraction form, using a slash or using negative indices. Prefixes should not be separated by a space.

TABLE 1.4.2 Examples of correct and incorrect use of symbols for derived units

Rule	✓ correct	✗ incorrect
Do not mix written numbers and symbols.	thirty grams 30 g	thirty g 30 grams
Do not put spaces between prefixes and unit symbols.	kg mL kPa	k g m L k Pa
Write the correct case for symbols (upper- or lower-case).	1 mm 84 kg 5.2 mL	1 Mm 84 KG 5.2 ml
SI units should not be followed by a full stop unless they are at the end of a sentence. The symbols for units are not abbreviations.	mL	m.L.
Only use upper-case letters for unit symbols named after people. The exception is 'L' for litre, to avoid confusion with 'l' and the numeral '1'. Only the symbol, not the name, uses upper-case.	J (the unit of energy is joule, named after James Joule) L mL	j Joule l Litre milliLitre
Units named after people can take the plural form by adding an 's' when used with numbers greater than one. Never do this with the unit symbols.	two kilojoules	2 kJs
The product of a number of units is shown by separating the symbol for each unit with a space.	0.1 mol dm^{-3}	0.1 moldm^{-3}
The division or ratio of two or more units can be shown in fraction form, using a slash, or using negative indices.	0.1 mol dm^{-3} $Jg^{-1}°C^{-1}$ kJ/s	0.1 moldm^{-3} $Jg^{-1}°C^{-1}$ kJs
When units are displayed with a negative exponent such as g mL^{-1}, the negative value of the exponent signifies the division of the units, or per unit. In this case, g mL^{-1} is the same as g/mL, or grams per millilitre.	1000 kJ mol^{-1} 13 °C/g	1000 kJ/mol^{-1} 13 °C/g^{-1}

Table 1.4.3 provides some examples of correct and incorrect use of symbols for derived units.

TABLE 1.4.3 Examples of the use of symbols for derived units

Correct symbol ✓	Incorrect symbol ✗
J $g^{-1}°C^{-1}$	$Jg^{-1}°C^{-1}$
g mol^{-1}	gmol^1
kPa	K Pa

When units are displayed with a negative exponent, such as mol L^{-1}, the negative value of the exponent signifies the division of the units; in other words, per unit. In this case, mol L^{-1} is the same as mol/L, or moles per litre.

Worked example 1.4.1

UNIT CONVERSIONS

Calculate how many litres are in one cubic kilometre.	
Thinking	**Working**
Write each unit in terms of m^3. Remember that a litre is the volume of a cube $0.1 \times 0.1 \times 0.1$ m Remember that a km^3 is a cube that has sides of 1000 m by 1000 m by 1000 m.	$1\,L = 0.001\,m^3$ and $1\,km^3 = 1000\,m \times 1000\,m \times 1000\,m$ $= 10^9\,m^3$
Take the inverse of each equation to find the equivalent volume in m^3.	$1m^3 = \frac{1}{10^9} = 10^{-9}\,km^3$ and $1m^3 = \frac{1}{0.001} = 10^3\,L$
Now equate them since they are both equal to $1\,m^3$.	$10^{-9}\,km^3 = 10^3\,L$ $\therefore \frac{10^3\,L}{10^{-9}\,km^3} = 10^{12}\,L/km^3$
State the answer.	There are 10^{12} or one trillion litres in 1 cubic kilometre.

➤ Try yourself 1.4.1

UNIT CONVERSIONS

If a person weighs 6.0×10^1 kg and the mass of a single carbon atom is 1.99×10^{-23} g, calculate how many carbon atoms would be needed to equal the mass of the person.

1.4 Review

SUMMARY

- Scientists use conventional ways of writing numerical measurements. The common standard is to use System International (SI) units.
- Units with a prefix should be converted into scientific notation before they are used in calculations.
- When converting units, it is very useful to write the conversions as fractions and then cross-cancel out to ensure that the conversion is done correctly.
- When units are displayed with a negative exponent (power of ten), the negative value of the exponent signifies the division of the units. For example, g mL^{-1} is the same as g/mL or grams per millilitre.
- When writing and recording measurements, remember:
  - use numerals for numbers that are accompanied by a symbol
  - numbers and symbols should not be mixed
  - do not put spaces between prefixes and unit symbols
  - always put a space between the last numeral and the unit symbol
  - write the correct case for symbols (upper- or lowercase).

KEY QUESTIONS

Describe

1 State the standard unit of measurement, and the symbol, for the following quantities.
- **a** time
- **b** temperature
- **c** length
- **d** mass

2 List the following units from largest to smallest: **centi**, milli, deci, mega, kilo, nano, micro.

Apply

3 Explain why it is important that general society, not just the scientific community, needs a systematic and consistent set of units for measurement.

Analyse

4 Convert 30.00 mL into L.

5 Identify any errors in the recordings below, and correct them.
- **a** 23 mL/g h^{-1}
- **b** 79 Kl s^{-1}
- **c** four litres
- **d** 1.56 grams

6 In some plants, the rate of photosynthesis is measured by measuring the change in atmospheric CO_2 in moles per m^2 of leaf area, per second (mol/m^2/s or mol $m^{-2}s^{-1}$). If a plant was measured to photosynthesise CO_2 at a rate of 25 umol $m^{-2}s^{-1}$, calculate the number of moles of CO_2 this plant would photosynthesise per second and per hour.

1.5 Measuring precision and accuracy using uncertainties and percentage error

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain the difference between accuracy and precision
- explain the difference between uncertainty and error
- write measurements with uncertainty and error values
- write measurements and calculations to the correct number of significant figures
- perform calculations using significant figures
- differentiate between accuracy and precision, and know how to express these quantitatively using significant figures
- explain the difference between mistakes, systematic uncertainties and random uncertainties
- perform calculations that contain uncertainty or error values
- understand that all measurements made in biology experiments are subject to a level of uncertainty.

All measurements are limited by the instrument used and the observer. Accuracy of the measurements and results is therefore affected. Instruments have a prescribed level of precision and cannot measure to less than the smallest increment of the device. In addition, a scientist's technique and experience with the instrument influences the precision with which the reading can be made. Environmental conditions, such as the ambient air temperature, can affect the instrument and the observer's accuracy with measurements.

Thus, in science it is impossible to measure a true value, as there will always be some degree of **uncertainty**. This results in **error** and uncertainty in the values measured or observed. Well-designed experiments can measure closer to the true value than others, and experimentation in closed systems enables more accurate and precise measurements. However, closed system measurements do not reflect true natural values.

Therefore, science can be thought of as developing evidence-based results that have some limitations. Measurements with uncertainty and errors are normal in science and so results are presented with a **range** (e.g. ±0.5) indicating possible precision or are processed through statistical calculations.

ACCURACY AND PRECISION

In science and statistics, the terms *accuracy* and *precision* have very specific and different meanings.

- **Accuracy** is the ability to obtain the correct measurement. To obtain accurate results, you must minimise **systematic errors**.
- **Precision** is the ability to consistently obtain the same measurement. To obtain precise results, you must minimise **random errors**.

To understand more clearly the difference between accuracy and precision, think about firing arrows at an archery target (Figure 1.5.1). Accuracy is being able to hit the bullseye, whereas precision is being able to hit the same spot every time you shoot.

Any measurement made in biology needs to be both accurate and precise.

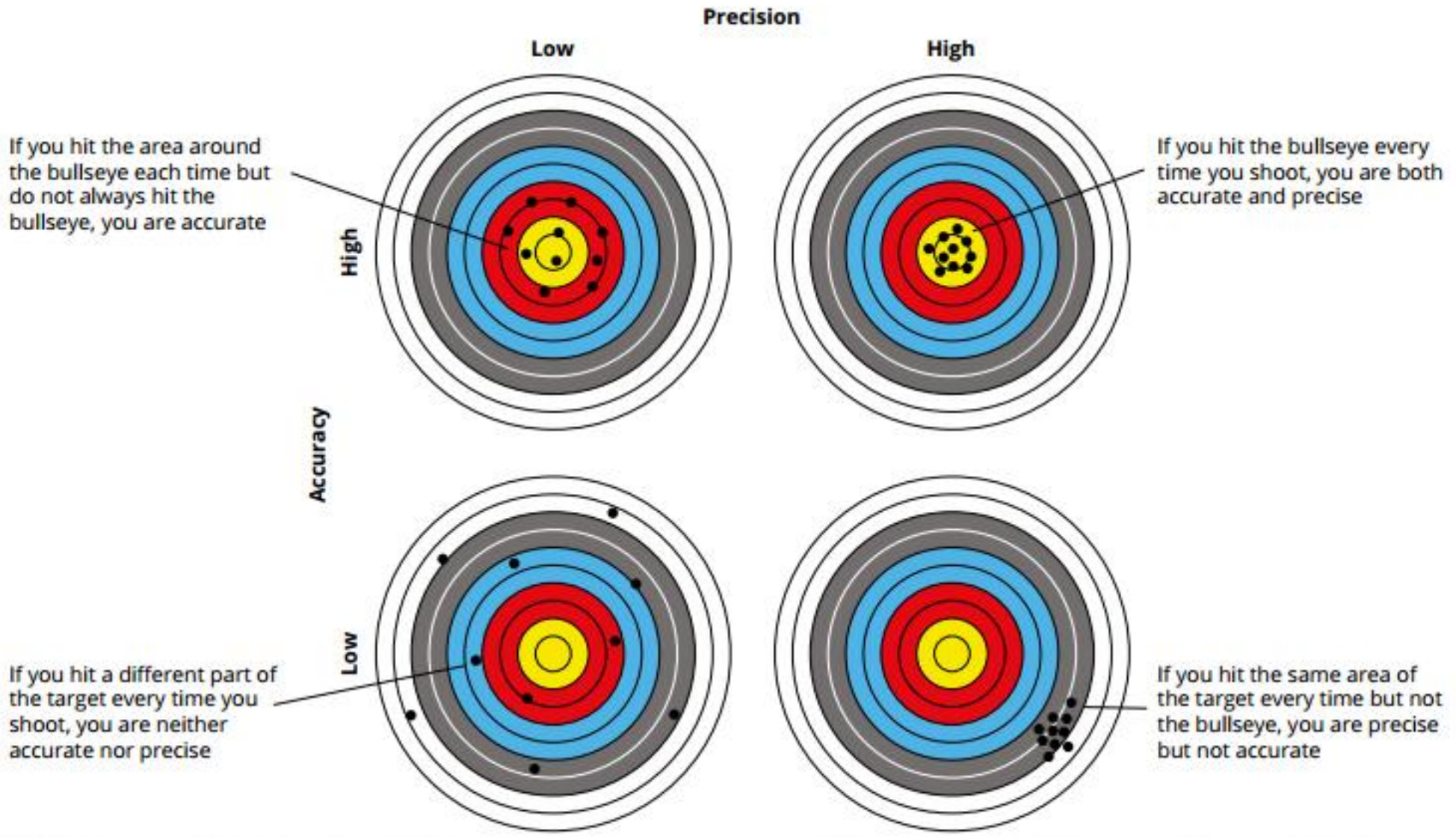

Precision
Low
High
Accuracy
High
Low
If you hit the area around the bullseye each time but do not always hit the bullseye, you are accurate
If you hit the bullseye every time you shoot, you are both accurate and precise
If you hit a different part of the target every time you shoot, you are neither accurate nor precise
If you hit the same area of the target every time but not the bullseye, you are precise but not accurate

FIGURE 1.5.1 Using a dartboard with darts illustrates the difference between accuracy and precision. In summary, measurements should be both precise and as accurate as you can make them.

FIGURE 1.5.2 A 10 mL graduated pipette is calibrated to deliver volumes to an accuracy of one-tenth of a millilitre, or 10 mL ± 0.1 mL. The pipette has major divisions of 1 mL and minor divisions of 0.1 mL. You can estimate to 0.05 mL and record volumes to 2 decimal places, for example, 3.80 mL or 4.55 mL.

Recording numerical data

When using instruments to measure data, the number of significant figures (or digits) and decimal places you use is determined by how precise your measurements are.

The overall precision of the measurement depends on the scale, accuracy and precision of the instrument and technique you are using (Figure 1.5.2). For example, a beaker may be used to measure volumes approximately and has limited accuracy of, for example, ±5%; however, a graduated pipette is much more accurate, with accuracies of ±0.1% or ±0.2%. In addition, your pipette may be accurate but if your technique using the pipette is variable, the overall accuracy and precision will be limited.

When you record **raw data** and report processed data, you must use the number of significant figures available from your equipment or **observation**. (This is discussed in more detail in Module 1.5.2) Using either a greater or smaller number of significant figures can be misleading. For example, Table 1.5.1 shows measurements of five tissue samples taken using an electronic balance accurate to 2 decimal places. The data was entered into a spreadsheet to calculate the mean, which was displayed with 4 decimal places. You would record the mean as 20.83 g, not 20.8260 g, because 2 decimal places is the precision limit of the instrument. Recording 20.8260 g would be an example of false precision.

TABLE 1.5.1 An example of false precision in a data calculation

Sample	1	2	3	4	5	Mean
Mass (g)	20.13	20.62	21.22	20.99	21.17	20.8260

UNCERTAINTY AND ERROR

Uncertainty is a range of measurements from the experiment, within which the true value is expected to lie. Due to variations, an estimation is made to suggest where the true value should be found among the measurements. Uncertainty is one indicator of a limitation. Expressed as a ± value, uncertainty can be derived either from the known precision of the equipment or alternatively from calculations involving the range of trials.

Error is the difference between a measurement and the true value. This is not an indication of human error or mistakes. It is due to an inability to achieve the true value. Error can be reduced but cannot be eliminated, and therefore it is important to understand and record the error and report it. Two types of errors can occur—systematic error and random error (Table 1.5.2).

TABLE 1.5.2 Systemic and random errors

Error	Definition	Cause	Example
systematic	An error in measurements caused by the design of a system (e.g. methodology) or instrument (uncalibrated), which results in the measurements shifting in a systematic direction. The mean may be displaced or varied in a predictable way affecting the accuracy of the measurement. It is quantified by calculating percentage error.	• instrument calibration • choice of instrument • precision of instrument (increments) • precision of instrument (sample rate—per second) • sampling (choice of population or representatives) • heat loss or gain with the surroundings	The balance used for weighing out samples reads 0.05 g too high for all your masses (because it was improperly tared (set to zero) at the start of the experiment). Consistently reading liquid volume measurements incorrectly from the top of the meniscus instead of the bottom. Error of parallax – reading a meter display consistently from an angle instead of directly in front.
random	An error that affects the measurement in an unpredictable way, resulting in an even variation (fluctuation) in measurements above and below the expected true value. This affects the precision of the measurement. It is quantified by calculating percentage uncertainty.	• estimation (measuring between increments) • sampling (frequency, spread, increments) • parallax • instrument sensitivity • noise	Using a universal indicator to determine the pH of a solution, in which the pH must be estimated by the shade and colour of the paper (e.g. yellow/orange versus bright orange). Measuring the time taken for a reaction to occur has a random error associated with the identification of when the reaction is judged to be complete. If a person's pulse was counted for 20 s and then the measurement was multiplied by three to determine beats per minute, this methodology allows the heart rate to change over 20 s while the pulse is measured. The measured heart rate will shift in an unpredictable way.

Uncertainties and errors should always be calculated or measured and then reported. They then form part of the Analysis and Interpretation (see Module 1.7 Data Analysis and Module 1.10 Finding) as well as the Evaluation in a report. Also, it is a good idea to consider uncertainty and error when planning experiments.

Calculating uncertainty in measurement

The goal when taking measurements is to obtain repeat values (trials) that closely agree. We expect repeat measurements to vary somewhat based on the precision of the equipment. For example, when using a measuring cylinder for the volume of solution, the **measurement uncertainty** of an analog instrument is half the smallest increment (for digital it is the smallest decimal place on the readout). Using the particular instrument, the true value is expected to fall within this range (e.g. measured value ± 0.15 mL, for a 150 mL volumetric cylinder).

$$\text{measurement uncertainty} = \pm \frac{\text{smallest increment}}{2}$$

When averaging repeat measurements, the uncertainty should be reported alongside your average. Known as **uncertainty of the mean**, it represents the precision of the method expressed as the range of experimentally obtained values. The following equation is a simple way to calculate the uncertainty of the mean.

$$\text{uncertainty of the mean} = \pm \frac{(\text{upper value} - \text{lowest value})}{2}$$

Worked example 1.5.1

CALCULATING UNCERTAINTY OF THE MEAN

An experiment was conducted to measure the length of time it takes to convert a substrate to a product in an enzymatic reaction. Three replications of the experiment produced the times 2.50, 3.48 and 2.81 s, the average time being 2.93 s. Calculate the uncertainty of the mean.

Thinking	Working
Write down the formula.	$\text{uncertainty} = \pm \frac{(\text{upper value} - \text{lowest value})}{2}$
Insert the values and then calculate.	$\text{uncertainty} = \pm \frac{(3.48 - 2.50)}{2}$ $= \pm 0.49 \text{ s}$
Combine measurement with uncertainty.	2.93 s ± 0.49 s

➤ Try yourself 1.5.1

CALCULATING UNCERTAINTY OF THE MEAN

Students measured the amount of oxygen produced by a plant over an hour. They performed the trial three times over three consecutive days. The results were 4.9, 5.5 and 5.2 mL. Calculate the uncertainty.

In experimental investigations where a number of trials are performed, you have the opportunity to describe the level of uncertainty in two numerical ways—measurement uncertainty and uncertainty of the mean. When these uncertainties are expressed in the same units as the value they describe, they are an **absolute uncertainty** ± (eg. 50cm ± 0.5cm).

Uncertainty can also be reported as a percentage of the measurement; this is known as **percentage uncertainty**. To calculate the percentage uncertainty, apply this formula:

$$\text{percentage uncertainty} = \pm \left(\frac{\text{absolute uncertainty}}{\text{measurement}} \right) \times 100$$

SKILLBUILDER 1.5.1

Calculating absolute measurement uncertainty for instrumental error

Calculate the absolute measurement uncertainty for instrumental error during an experiment where the temperature was measured at 32°C using a glass thermometer with increments of 1°C and also a digital thermometer with increments of 0.1°C.

For the glass thermometer

1 Write down the formula.

$$\text{uncertainty} = \pm\frac{\text{smallest increment}}{2}$$

2 Insert the values and calculate.

$$\text{uncertainty} = \pm\frac{1}{2}$$
$$= \pm 0.5°C$$

3 Combine the measurement with the uncertainty.

$32°C \pm 0.5°C$

For the digital thermometer

For digital instruments, the absolute uncertainty is equal to the smallest decimal place displayed.

1 Write down the formula.

uncertainty = smallest increment

2 Insert the values and calculate.

uncertainty = 0.1

3 Combine the measurement with the uncertainty.

$32.0°C \pm 0.1°C$

Worked Example 1.5.2

CALCULATING PERCENTAGE MEASUREMENT UNCERTAINTY FOR INSTRUMENTAL ERROR

The temperature during an experiment was measured and then reported as at $32.00°C \pm 0.05°C$. Calculate the percentage measurement uncertainty.

Thinking	Working
Write the formula for percentage uncertainty.	$\text{percentage uncertainty} = \pm\left(\frac{\text{absolute uncertainty}}{\text{measurement}}\right)\times 100$
Insert the values and calculate.	$\text{percentage uncertainty} = \pm\left(\frac{0.05}{32}\right)\times 100$ $= 0.16\ \%$
Combine the measurement with the percentage uncertainty and express to the appropriate number of significant figures.	$32.00°C \pm 0.16\%$

➤ Try yourself 1.5.2

CALCULATING PERCENTAGE MEASUREMENT UNCERTAINTY FOR INSTRUMENTAL ERROR

The pH of a solution was determined to be 11.4 ± 0.6. Calculate the percentage measurement uncertainty.

Calculating percentage error in values

As stated above, scientist try to calculate the 'true' or 'correct' value. However, due to equipment and/or method, experimental results may give an inaccurate value. The density of water is generally stated to be 1 kg m^{-3}; however, the exact density at 4.0 °C is 0.9998395 kg m^{-3}.

The percentage error between the measured value and the true value provides information about validity and systematic error (see Part B) and can be calculated using the percentage error formula:

$$\text{percentage error (\%)} = \left|\frac{\text{measured value} - \text{true value}}{\text{true valued}}\right| \times \frac{100}{1}$$

In this case, the measured value is replaced with the accepted value. The percentage error in the accepted (rounded) value of 1 kg m^{-3} is less than 0.02 %. This indicates that using 1 kg m^{-3} is acceptable since the percentage error is very small and likely to be much smaller than experimental uncertainty.

Worked example 1.5.3

CALCULATING PERCENTAGE ERROR

Calculate the percentage error between the true density of water at 4.0°C (0.9998395 kg m^{-3}) and the generally accepted (rounded) value of 1 kg m^{-3}.

Thinking	Working
Write down the formula.	$\text{percentage error (\%)} = \left\|\frac{\text{measured value} - \text{true value}}{\text{true value}}\right\| \times \frac{100}{1}$
Insert the values then calculate.	$\text{percentage error (\%)} = \left\|\frac{1 - 0.9998395}{0.9998395}\right\| \times \frac{100}{1}$ $= \frac{0.00016505}{0.9998395} \times \frac{100}{1}$ $= 0.01605\ \%$

➤ Try yourself 1.5.3

CALCULATING PERCENTAGE ERROR

The laboratory technician made up a standard solution of 0.200 M. A student calculated the concentration of the solution to be 0.185 M. Calculate the percentage error.

Significant figures

When measuring phenomena during experimentation, the precision of the instruments determines the number of significant figures and decimal places you record in the data.

Significant figures convey meaning and precision and the number of significant figures used depends on the scale of the instrument. You should record data using the number of significant figures available from the equipment or observation. Using either a greater or smaller number of significant figures is misleading.

When writing an answer to the correct number of significant figures, you may need to use rounding if the initial answer has more figures (digits) than you need. These extra figures are called 'non-significant figures'. If the first non-significant figure is ≥5, round up the number. If the first non-significant figure is <5, then do not round up. For example, if the initial answer for a calculation was 2.1259 but you only needed an answer to 3 significant figures, you would round it to 2.13. If the initial answer was 2.1241, then to 3 significant figures, this becomes 2.12.

SKILLBUILDER 1.5.2

Identifying significant figures

When giving an answer to a calculation, it is important to take note of the number of significant figures that you use.

You should give an answer that is as accurate as possible. However, an answer cannot be more accurate than the data or the measuring device used to calculate it. For example, if an electronic balance that measures to the nearest gram shows that an object has a mass of 56g, then the mass should be recorded as 56g, not 56.0g. This is it only measures to the whole gram so you do not know if it is 56.0g, or 56.1g, or 56.2g or 55.8g.

The number 56 has 2 significant figures. Recording to 3 significant figures (e.g. 56.0g or 55.8g) would not be scientifically 'honest'.

If this mass of 56g is used to calculate another value, it would also not be 'honest' to give an answer that has more than 2 significant figures.

Determining the number of significant figures for an answer depends on the kind of calculation you are doing.

- If you are multiplying or dividing, use the smallest number of significant figures provided in the initial values.
- If you are adding or subtracting, use the smallest number of decimal places provided in the initial values.

Working out the number of significant figures

Use the following rules to avoid confusion in determining how many significant figures are in a number.

1. All non-zero digits are always significant; for example, 21.7 has 3 significant figures.
2. All zeros between two non-zero digits are significant; for example, 3015 has 4 significant figures.
3. A zero to the right of a decimal point and following a non-zero digit is significant; for example, 0.5700 has 4 significant figures.
4. Any other zero is not significant, as it will be used only for locating decimal places; for example, 0.005 has just 1 significant figure with the leading zero and the two zeros after the decimal point not significant, they just place the decimal point in the correct position.

Worked example 1.5.4

IDENTIFYING SIGNIFICANT FIGURES

a Identify which of the following is written to 2 significant figures.

A 30.1
B 0.000 40
C 0.5
D 5.12

b George multiplied 1.22 by 1.364. Identify which of the options below shows the result of this multiplication with the correct number of significant figures.

A 1.66
B 1.664
C 1.65
D 1.7

Thinking	Working
a The zeros that come before the integer 4 are not significant, whereas those that follow the integer are significant.	B is correct. A has 3 significant figures. C has 1 significant figure. D has 3 significant figures.
b 1.22×1.364 $= 1.66408$ Answer is to have 3 significant figures $= 1.66$	A is correct. When two numbers are multiplied, use the smallest number of significant figures in the initial values to report your answer. In George's multiplication, the answer is 1.66408, but since 1.22 has 3 significant figures, the correct answer is 1.66 with 3 significant figures.

➤ Try yourself 1.5.4

IDENTIFYING SIGNIFICANT FIGURES

Identify which of the following shows 41 written to 4 significant figures.

A 0.410
B 4.100
C 41.00
D 4100

Worked example 1.5.5

ACCURATELY RECORDING MEASUREMENTS FROM INSTRUMENTS

When measuring the amount of solution for an acid/base experiment, you need to record the results accurately and be able to identify the appropriate number of significant figures to use.	
a State the measurement shown on this plastic pipette, and identify the number of significant figures in this result. 	**b** State the measurement shown on this glass pipette, and identify the number of significant figures in this result.
Thinking	**Working**
a The increments shown on the plastic pipette determine the accuracy with which the result can be recorded. If you add extra numbers to the result, it becomes inaccurate.	The plastic pipette is marked with increments of 0.05 mL. Therefore all results must be recorded to the nearest 0.01 mL.
Determine where the meniscus (the lowest point) of the liquid falls.	The meniscus of the liquid in the pipette is closest to the 2.00 mL marking, therefore the result must be recorded as 2.00 mL.
Determine the number of significant figures included in this result.	The measurement of 2.00 mL has 3 significant figures.
b The increments shown on the glass pipette determine the accuracy with which the result can be recorded. If you add extra numbers to the result, it becomes inaccurate.	The glass pipette is marked with increments of 0.5 mL. Therefore all results must be recorded to the nearest 0.01 mL.
Determine where the meniscus (the lowest point) of the liquid falls.	The meniscus of the liquid in the pipette is closest to the 2.00 mL marking, therefore the result must be recorded as 2.00 mL.
Determine the number of significant figures included in this result.	The measurement of 2.00 mL has 3 significant figures.

➤ Try yourself 1.5.5

ACCURATELY RECORDING MEASUREMENTS FROM INSTRUMENTS

When measuring the temperature for an experiment, you need to record the results accurately and be able to identify the appropriate number of significant figures to use.

a State the measurement shown on this glass alcohol thermometer and identify the number of significant figures in this result.

b State the measurement shown on this digital thermometer and identify the number of significant figures in this result.

FIGURE 1.5.3 This 5 mL plastic pipette has 1 mL increments, so measurement values are recorded to one significant figure.

Significant figures are the exact digits observed when recording the results of an observation or experiment. For example, if the volume of O_2 gas produced and measured from a plant photosynthesising during a 6-hour experiment was 6.0 mL then the recorded measurement is 6.0 mL with two significant figures, not 6 mL. This is because exactly 6.0 mL was observed.

If you measured 2 mL with the pipette in Figure 1.5.3, then you should record 2 mL, not 2.0 mL. This is because the measurement was not precise to one-tenth (0.1) of a millilitre. Using this instrument, the value is measured and recorded to only one significant figure.

Table 1.5.3 shows some examples of measurements and how many significant figures they have.

TABLE 1.5.3 Measurements and significant figures

Measurement	Number of significant figures
15	2
3.5	2
3.50	3
0.037	2
1401	4
1400	2

Using either a greater or smaller number of significant figures can be misleading. For example, Table 1.5.4 shows three measurements to 2 decimal places (4 significant figures). The data was entered into a spreadsheet to calculate the **mean**, which was displayed with 3 decimal places. You would record the mean as 19.52 mL, not 19.517 mL, because 2 decimal places is the precision limit of the instrument. Recording 19.517 g would be an example of false precision.

TABLE 1.5.4 An example of false precision in a data calculation

Trial	1	2	3	Mean
Volume (mL)	19.45	19.55	19.55	19.517

> It is important to record the exact numbers measured during an experiment. This determines the significant figures, and later the accuracy and precision of the analysis. If a zero is observed, record it, as it is the exact amount measured. This includes all zeros before and after decimal places.

Determining an uncertainty in a single measurement

When using an instrument to measure a property of an object, there will always be a limit as to how precise the reading can be.

The precision that can be read off a single instrument is called the limit of reading. This is the smallest visible graduation on an analog instrument (see Figure 1.5.3), or if a digital instrument is used, then the limit of reading is usually the smallest decimal place given on the screen.

For most analog instruments, the **absolute uncertainty** in a measurement is exactly half of the limit of reading for each part of the measured object that has to be lined up. This means that the actual measurement could be anywhere from *half* of the smallest graduation too big to *half* of the smallest graduation too small.

Some analog instruments have a zero mark and a reading mark, such as a ruler shown in Figure 1.5.4a. The object to be measured has to be lined up at *both* ends, thus doubling the absolute uncertainty. For instance, when using rulers or burettes, the absolute uncertainty is equal to the limit of reading, rather than half of it.

In a digital instrument the limit of reading is the smallest decimal place given on the screen. An example of this is a high-precision electronic balance as shown in Figure 1.5.4b.

Usually, the absolute measurement uncertainty in a measurement will have one significant figure, or two significant figures if the first digit is 1. This is done because the uncertainty of a measurement is meant to be an estimate and writing more than two significant figures is therefore meaningless.

When the absolute uncertainty has been determined, the measurement should be rounded so that it and the absolute uncertainty have the same number of *decimal places*. This is to keep the measurement and its uncertainty to the same precision.

Examples of measurements and their absolute uncertainties are shown in Table 1.5.5.

a

b

FIGURE 1.5.4 (a) A ruler with a limit of reading of 1 millimetre, (b) an electronic balance with a limit of reading of 0.001 g (because there are 3 zero places after the decimal point displayed on the screen)

TABLE 1.5.5 Examples of writing a measurement and its absolute uncertainty

Measurement and absolute uncertainty	Comments
48.2 ± 1.0°C	The absolute uncertainty is written to 2 significant figures because the first digit is 1. The measurement and uncertainty then have the same number of decimal places.
99.99 ± 0.01 s	The measurement and absolute uncertainty are written to 2 decimal places, with one significant figure for the uncertainty. This measurement of time may have been recorded using a digital device, such as a data logger, since the precision is quite high.
0.000000031 ± 0.000000005 kg	This is technically a correct way of writing a measurement and its absolute uncertainty, but it is normally written in scientific notation so that the measurement and uncertainty have the same power of ten, i.e. $(3.1 \pm 0.5) \times 10^{-8}$ kg

The limit of reading and absolute uncertainties of common equipment used in a science laboratory are shown in Table 1.5.6.

TABLE 1.5.6 A summary of common instruments and their uncertainties

Instrument	analog or digital	Limit of reading	Absolute uncertainty	Example	Notes
mm ruler	analog	1 mm	± 1.0 mm	123.8 ± 1.0 mm	Half of the limit of reading is 0.50 mm, but since the object to be measured has to be lined up at both ends, the uncertainty is double this, or ±1.0 mm.
cm ruler	analog	1 cm	± 1.0 cm	31.4 ± 1.0 cm	Half of the limit of reading is 0.50 cm, but since the object to be measured has to be lined up at both ends, the uncertainty is double this, or ±1.0 cm.
thermometer	analog	1.0 °C	± 0.5 °C	24.4 ± 0.5 °C	Only the top part of the liquid in the thermometer has an uncertainty, so the uncertainty is exactly half of the limit of reading.
measuring cylinder	analog	1.0 mL	± 0.5 mL	15.7 ± 0.5 mL	The limit of reading and absolute uncertainty depend on the size of the measuring cylinder.
burette	analog	0.1 mL	± 0.05 mL	20.10 ± 0.05 mL	Half of the limit of reading is 0.05 mL, but since the object to be measured has to be lined up at both ends, the uncertainty is double this, or ±0.1 mL.
volumetric pipette	analog	varies	varies ± 0.03 mL	25.00 ± 0.03 mL	The uncertainty for the pipette is based on the precision given on the pipette. The precision varies based on the size and quality of the pipette.
stopwatch	digital	0.01 s	from ± 0.1 to ± 0.4 s	9.6 ± 0.2 s	The limit of reading is not the absolute uncertainty for a stopwatch because the user's reaction time (usually between 0.1 and 0.4 s) needs to be taken into consideration.
electronic balance	digital	typically 0.01 g	± 0.01 g	7.03 ± 0.01 g	The limit of reading is usually the smallest decimal place given on the screen.

For analog instruments that have one end on the scale to line up (such as a measuring cylinder, thermometer or voltmeter), the absolute measurement uncertainty is half of the smallest increment scale, or limit of reading.

For analog instruments that have two ends on the scale to line up (such as rulers and burettes), the absolute measurement uncertainty is equal to the smallest increments on the scale, or limit of reading.

For digital instruments, the absolute measurement uncertainty is equal to the smallest decimal place displayed.

The absolute uncertainty should be written to 1 or a maximum of 2 significant figures.

These steps will help you write a measurement and its absolute uncertainty.

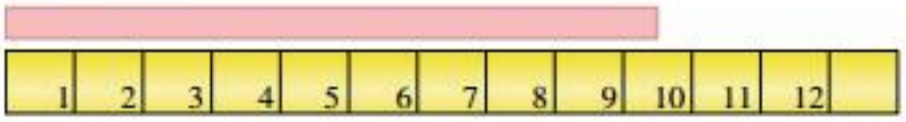
1. Determine the absolute uncertainty from the type of instrument used to measure the quantity.
2. Measure the quantity, including an estimate of the final significant figure.
3. If the first digit of the absolute uncertainty is 1, then one more significant figure can be quoted. Usually this extra digit is zero.
4. The measurement is then rounded up or down to the same number of decimal places, or place value holders, as the uncertainty; however, the uncertainty should not be presented to more than 2 significant figures.
5. Do not forget to include the unit of measurement for the quantity.

Worked example 1.5.6

READING MEASUREMENTS

For each of the following objects, determine its measurement and uncertainty.

a the length of the pink rectangle measured with a ruler calibrated in centimetres



b the temperature of tap water measured using an analog thermometer

c the temperature of tap water measured using a digital thermometer

Thinking	Working
a The pink rectangle is between 9 cm and 10 cm in length, so the best estimate for this is 9.6 cm.	length, $L = 9.6\,\text{cm}$
The limit of reading is 1.0 cm, so this is the uncertainty in a centimetre ruler.	$\Delta L = \pm 1.0\,\text{cm}$
Write the measurement and uncertainty.	length = 9.6 ± 1.0 cm
b The temperature is between 20°C and 21°C, with the best estimate of 20.2°C.	temperature, $T = 20.2\,^{\circ}\text{C}$
The limit of reading is 1.0°C, so the uncertainty is half of this, i.e. 0.5°C.	$\Delta T = \pm 0.5\,^{\circ}\text{C}$
Write the measurement and uncertainty.	temperature = 20.2 ± 0.5°C
c This temperature measurement is done using a digital thermometer, so the temperature is simply read off the screen at 31.2°C.	temperature, $T = 31.2\,^{\circ}\text{C}$
The uncertainty in this measurement is the smallest decimal value, i.e. ± 0.1°C.	$\Delta T = \pm 0.1\,^{\circ}\text{C}$
Write the measurement and uncertainty.	temperature = 31.2 ± 0.1°C

➤ Try yourself 1.5.6

READING MEASUREMENTS

For each of the following objects, determine its measurement and uncertainty.

a the length of the pink rectangle measured with a ruler calibrated in centimetres

b a red liquid in an analog measuring cylinder

c the mass of this doughnut measured on an electronic balance

Calculations with uncertainties

There are a few rules when conducting calculations with numbers (or measurements) that have uncertainties. It is important to record the correct uncertainty once all the calculations are done so that the appropriate accuracy is reported. Table 1.5.7 outlines the rules for undertaking calculations using mathematical operations with data and its associated uncertainty.

Keep in mind that the rules for significant figures apply to all numerical reporting, including the uncertainties.

TABLE 1.5.7 Rules for calculations involving uncertainty

Calculation	Rule	Examples
addition and subtraction	add the absolute uncertainties	$5.4 \pm 0.05 + 3.2 \pm 0.05 = 8.6 \pm 0.1$ $5.4 \pm 0.05 + 3.2 \pm 0.1 = 8.6 \pm 0.15$ $5.4 \pm 0.05 - 3.2 \pm 0.05 = 2.2 \pm 0.1$ $5.4 \pm 0.05 - 3.2 \pm 0.1 = 2.2 \pm 0.15$
multiplication and division	add the percentage uncertainties	$3.0 \pm 0.05 \times 1.5 \pm 0.05$ $= 3.0 \pm 1.6\% \times 1.5 \pm 3.3\%$ $= 4.5 \pm 4.9\%$ $3.0 \pm 0.05 \div 1.5 \pm 0.05$ $= 3.0 \pm 1.6\% \div 1.5 \pm 3.3\%$ $= 2.0 \pm 4.9\%$
multiplying by a constant (absolute uncertainty)	multiply the uncertainty	$3.14 \times (4.1 \pm 0.05)$ $= 12.9 \pm 0.16$
multiplying by a constant (percentage uncertainty)	maintain the uncertainty	$3.14 \times (4.1 \pm 1.6\%)$ $= 12.9 \pm 1.6\%$

Uncertainty due to nature

In nature, there are many sources of variation that make it impossible to measure the same value twice. Examples of variations in nature are shown in Figure 1.5.5. They include:

- genetic differences
  - variation in the genetic code
  - variation in the regulation of the code
- intracellular differences in molecular processes and capacity
- extracellular differences between cells, tissues, organs and systems in their environments
- differences in how organisms relate (intraspecies and interspecies relationships)
- various abiotic factors affecting biotic factors in habitats and ecosystems
- local, regional and global differences in ecological relationships.

There are a few terms used to describe the variations found naturally in biology, including 'genetic variation', 'biological variation' and 'natural variation'. Genetic variation is found in biological species and their populations. Therefore, when the term 'natural variation' is used, it often refers to genetic variation. When reporting natural, biological or genetic variation, it is best to choose one term and use it consistently.

The difficulty with uncertainty due to nature is that the true value can never be known. Biological experiments often are open systems, so are influenced by many variables. Also, many biological experiments measure non-invasive or indirect results, which may make it more difficult to achieve accuracy. For example, satellite imagery can be used to calculate distribution and abundance of an introduced plant species, like buffel grass, over a large area but may not be accurate at discriminating between all grass species on the ground in that area, especially during different seasons.

Measuring a range of values is still expected in biology; therefore, analysis and evaluation using uncertainty to acknowledge accuracy and precision is important when you interpret the results. Statistical analysis helps to analyse the data and range of measurements in order to determine uncertainty (see details earlier and Module 1.5 page e47 Review Summary).

FIGURE 1.5.5 There are many sources of natural variation when gathering biological data. (a) Close inspection of bees reveals variation in shades of colour and banding, which could be due to genetics and/or environment. (b) The same species of plant can be affected by different pH values in the soil, displaying variation within a habitat not due to genetics. (c) Genetic variation within a family. (d) Genetic variation within plants shown by wrinkled peas (left) and round peas (right).

1.5 Review

SUMMARY

- Accuracy is the ability to obtain the correct measurement. To obtain accurate results, you must minimise systematic errors.
- Precision is the ability to consistently obtain the same measurement. To obtain precise results, you must minimise random errors.
- Uncertainty is a range of measurements from the experiment within which the true value is thought to be.
- Error is the difference between a measurement and the true value; this is not an indication of human error or a mistake.
- Uncertainty values are presented as a range (e.g. ±0.3).
- A simple way to calculate the uncertainty of the mean is:

$$\text{uncertainty} = \pm\frac{(\text{upper value} - \text{lowest value})}{2}$$

- A simple way to calculate the measurement uncertainty is:

$$\text{measurement uncertainty} = \pm\frac{\text{smallest increment}}{2}$$

- For digital instruments, the absolute uncertainty is equal to the smallest decimal place displayed.
- The percentage uncertainty can be calculated using this formula:

$$\text{percentage uncertainty} = \pm\left(\frac{\text{absolute uncertainty}}{\text{measurement}}\right)\times 100$$

- The percentage error, when the experimental result can be compared with a theoretical or accepted result (value) can be calculated using this formula:

$$\text{percentage error (\%)} = \left|\frac{\text{measured} - \text{true value}}{\text{true value}}\right| \times \frac{100}{1}$$

- Discriminate between absolute uncertainty and percentage error.
- The rules for mathematical calculations involving uncertainty are listed in Table 1.5.5.
- Significant figures are a way of communicating how precise a measurement is.
- The number of significant figures used depends on the scale of the instrument.
- There are four specific rules governing how significant figures are written in any measurement. These are demonstrated in Skillbuilder 1.5.5.
  - All non-zero digits are significant.
  - All zeros between non-zero digits are significant.
  - Trailing zeros (either decimal or not) if measured are significant.
  - Leading zeros are not significant (in numbers with decimals, they only inform where the decimal is placed).
- All measurements made in science experiments are subject to a level of uncertainty or doubt to the precision of that measurement.
- Biological science in particular has natural sources of variation that add uncertainty to true values. Measuring a range of values and determining uncertainty is still expected.
- Systematic errors arise due to mistakes in calibrating or using equipment the wrong way for every measurement.
- Random errors are naturally occurring and cannot be eliminated.
- The limit of reading of an instrument is the smallest graduation on the device.
- When adding or subtracting measurements, the individual absolute uncertainties always add.
- When multiplying or dividing measurements, the individual percentage uncertainties always add.
- When raising a measurement to a power, the percentage uncertainty is multiplied by the absolute value of the power.
- When using measurements in a formula, perform any addition and subtraction operations first, followed by multiplication, division and indices.

1.5 Review *continued*

KEY QUESTIONS

Describe

1 Describe how values of uncertainty and error are presented.

2 Explain the difference between accuracy and precision.

3 Explain the difference between raw and processed data.

4 State the absolute uncertainty of the following instruments.
- **a** a cm ruler
- **b** a protractor graduated in degree one degree intervals
- **c** an electronic balance giving a reading to the nearest 0.001 g
- **d** an analog needle voltmeter graduated in 10 volt intervals

Apply

5 Identify the following scenarios as examples of random or systematic error.
- **a** You weigh the mass of a powder three times, using the same scales and get three different readings.
- **b** The tape measure you are using to measure distance has been stretched.
- **c** When electronic scales are zeroed, they read 0.05 g.

6 Explain why it would be unreasonable to state that the absolute uncertainty in a hand-controlled stopwatch is ±half of the limit of reading or ±0.005 s.

Analyse

7 Calculate the average and uncertainty of the mean of the two sets of data below.

Dataset A: 11.4, 10.9, 11.8, 10.6, 11.5, 11.1

Dataset B: 25.3, 27.4, 22.0, 26.1, 28.4, 23.1

8 Determine the percentage measurement uncertainty of the following instruments.
- **a** a garden pH meter graduated in 0.2 intervals
- **b** a burette graduated in 0.1 mL intervals
- **c** an electronic balance giving a reading to the nearest 0.001 g

1.6 Tables and graphing

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- create tables to record and organise data
- present data from tables into graphs
- distinguish between types of graphs
- select appropriate graphs to represent data
- represent missing data in graphs
- explain what an outlier is and know how to represent them in graphs.
- add error bars to data points on a graph.

PRESENTING DATA IN TABLES

Tables record number values and allow you to organise your data. In general, tables provide more detailed data than graphs, but it is easier to observe trends and patterns in data when they are in graph format than in table format.

Presenting raw data in tables

Tables organise data into rows and columns and can vary in complexity according to the nature of your data. Tables can be used to organise raw data and **processed data**, or to summarise results.

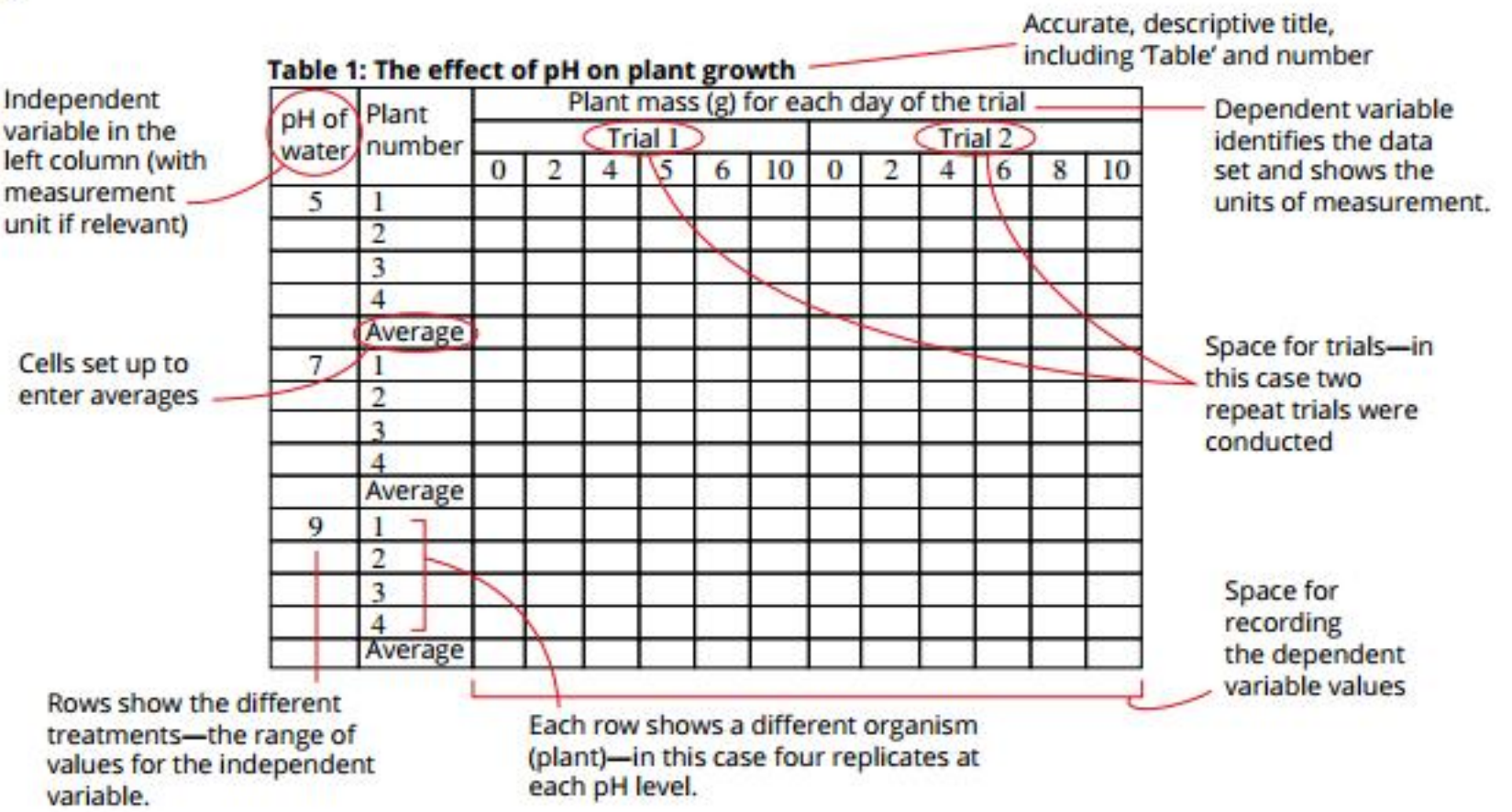
The simplest form of a table is a two-column or two-row chart. The first column (or top row) should contain the independent variable (IV – the one you determine and manage) and the following column(s) or row(s) should contain the dependent variable (DV – the outcome or results you measure in response to changes in the IV). When there are repeat experiments, or a series of trials, extra columns or rows are used for DVs, and a space created on the table for an average.

As you can see in Figure 1.6.1 (page e50), tables should have the following features:

- a descriptive title, with a table number if the report has more than one table of data
- column headings (including the units)
- aligned figures (align the decimal points)
- most often data should be entered with the same number of significant figures
- the independent variable placed in the left column or top row
- the dependent variables placed in the right column(s)
- numbers only in each cell, no units (ensure all values entered have the same units as the heading).

All tables presented in this student book use the features listed above and are useful models.

a


Accurate, descriptive title, including 'Table' and number
Table 1: The effect of pH on plant growth
Independent variable in the left column (with measurement unit if relevant)
pH of water
Plant number
Plant mass (g) for each day of the trial
Trial 1
Trial 2
0 2 4 5 6 10 0 2 4 6 8 10
Dependent variable identifies the data set and shows the units of measurement.
5 1
2
3
4
Average
Cells set up to enter averages
7 1
2
3
4
Average
Space for trials—in this case two repeat trials were conducted
9 1
2
3
4
Average
Space for recording the dependent variable values
Rows show the different treatments—the range of values for the independent variable.
Each row shows a different organism (plant)—in this case four replicates at each pH level.

b

Data entry correct

Treatment	Plant height (mm)
fertiliser	132
no fertiliser	119

c

Data entry incorrect (units go in the heading only)

Treatment	Plant height (mm)
fertiliser	~~132 mm~~
no fertiliser	~~119 mm~~

FIGURE 1.6.1 (a) Features of a good science table to enter raw data with spaces allowed for averages (b) Correct data entry (c) Incorrect data entry

Presenting processed data in tables

Table 1.6.1 shows the relationship between temperature and mean transpiration rate of a plant. It displays transpiration data in a processed format, because it displays the average of several values for each temperature. To report processed data accurately the uncertainty is also displayed, which will be a requirement for assessments. Again, units should only be entered for the headings.

TABLE 1.6.1 Effect of temperature on average transpiration rate, including uncertainty

Temperature (°C)	Mean transpiration rate ($mL\,g^{-1}\,h^{-1}$)
15	0.038 ± 0.002
25	0.043 ± 0.001
35	0.059 ± 0.001
45	0.074 ± 0.0015

PRESENTING DATA IN GRAPHS

A **graph** is a very effective tool for presenting data. Types of graphs include line graphs, pie charts, bar and column graphs. The best one to use will depend on the nature of the data and sometimes on the target audience. With practice you will develop the skill to select the best type of graph to use.

Scatterplots are used for continuous and discrete data. They are used to show the relationship between two variables when one variable is dependent on the other. IV is on the *x*-axis and DV is on the *y*-axis. The data is plotted on the graph as a series of points in pencil using a cross or small circle for each point.

A line graph is usually the best way to represent continuous quantitative data on a scatterplot. It may be appropriate to ruler-draw lines joining one point to the next (Figure 1.6.2) or to draw a single **line of best fit**, also known as a **trend line**. Note that a trend line can be either straight or curved (drawn freehand), whichever best fits the data points (Figure 1.6.3).

Continuous data exists and varies between the recorded points e.g. if time is plotted for 5 and 10 years, it also exists at 6, 7, 8, 9 years, even if not plotted.

Discrete data has distinct and separate values with no values between the recorded points e.g. the number of students in a classroom.

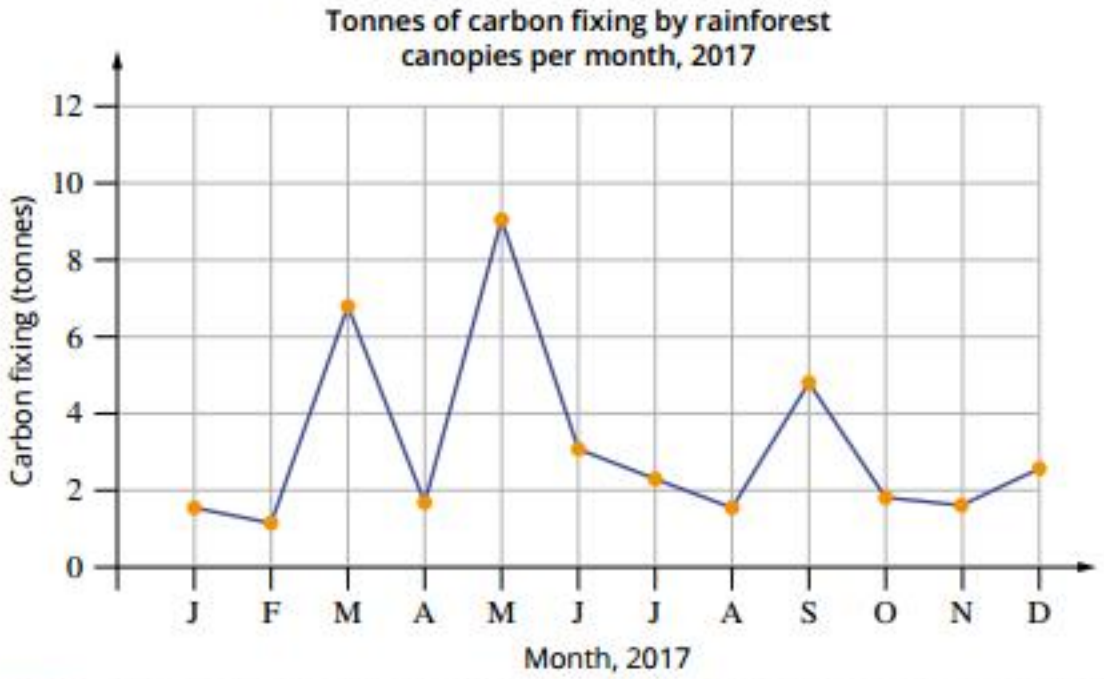


FIGURE 1.6.2 A line graph showing tonnes of carbon fixing by rainforest canopies per month in 2017, with lines ruled from each point to the next. For other sets of data, a line of best fit through the plotted points will be more appropriate (as in Figures 1.6.3 and 1.6.5)

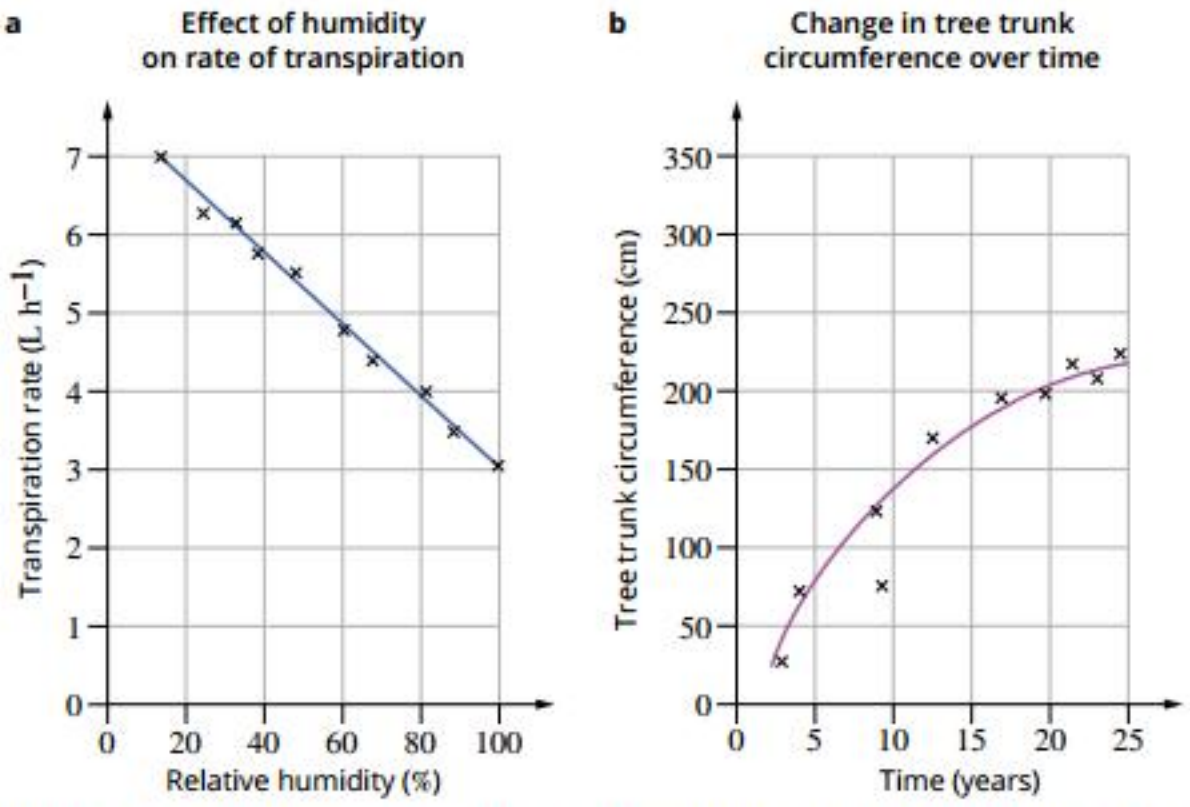


FIGURE 1.6.3 Scatterplot graphs showing (a) straight and (b) curved trend lines (lines of best fit)

Plotting data in graphs has many advantages.

- Data can be represented visually.
- Data points that are possibly incorrect (known as outliers) can be identified and a decision made if they can be ignored. Figure 1.6.3b has an example.
- Constants can be determined from the properties of the line or curve, such as the gradient.
- Uncertainties can be represented as **error bars** (Figure 1.6.4).
- A general trend **line of best fit** that matches the majority of the data points can be determined (Figure 1.6.5).
- Any known relationship between the dependent and independent variables can be supported or rejected, especially by the use of uncertainties.

Tables often provide more detailed data than graphs, but graphs can suggest a possible relationship between the dependent and independent variables more clearly, or highlight if none exists. From the shape of the curve, it is easier to observe trends, patterns and gaps in data in graphical form than in tabular form. For example, in Figure 1.6.3a the direct relationship between declining transpiration rate and increasing humidity is very obvious. In Figure 1.6.3b an outlier has been ignored in drawing the line of best fit. It can be seen that the DV (tree trunk circumference) increased steadily for the first 15 years then slowed in growth over the next 25 years, possibly trending to a plateau with little or no further increase. This might drive decisions for forestry scientists of when to harvest plantation trees, or when a plantation needs to be fertilised.

DRAWING GRAPHS IN BIOLOGY

Every graph used in biology must have the following features.

1 The graph must have a suitable set of axes for the independent and dependent variables.

Most biology graphs are only drawn in the first quadrant, but some graphs use all four quadrants (i.e. have negative values for the independent and/or dependent variables).

Remember that:

- the independent variable is the variable that you deliberately change (pre-set/manage) and always goes along the *x*-axis
- the dependent variable is the variable that changes (usually data collected as experimental results) and always goes on the *y*-axis.

2 The graph must have a suitable scale that allows information to be seen easily and accurately. For most graphs, it is important to choose a scale that is linear and has tick marks at common intervals (e.g. 2, 5, 10, 100 units).

3 The title and axes must be named. Make sure that each axis label shows information about the quantity and units. Give a descriptive title that is not just a repeat of the labels on the axes.

4 Data points should be plotted as a small circle or small cross indicating their position.

5 Error bars should be drawn where possible for each point. These indicate the absolute uncertainties in the independent (a horizontal line) and dependent (a vertical line) values. They are drawn as a horizontal line centred on the data point indicating the independent variable's uncertainty left and right of the value, and as a vertical line centred on the data point above and below the value, indicating the dependent variable's uncertainty. Combining the *x* and *y* error bars forms an error rectangle that the true measurement is located within (Figure 1.6.4). The smaller the errors bars for a given point, the more precise the measurement is.

i Linear scale on an axis means equal distances on the scale represent equal differences in value. For example, you must not jump from intervals of 10 units each to the next interval in sequence being a different value like 20 units.

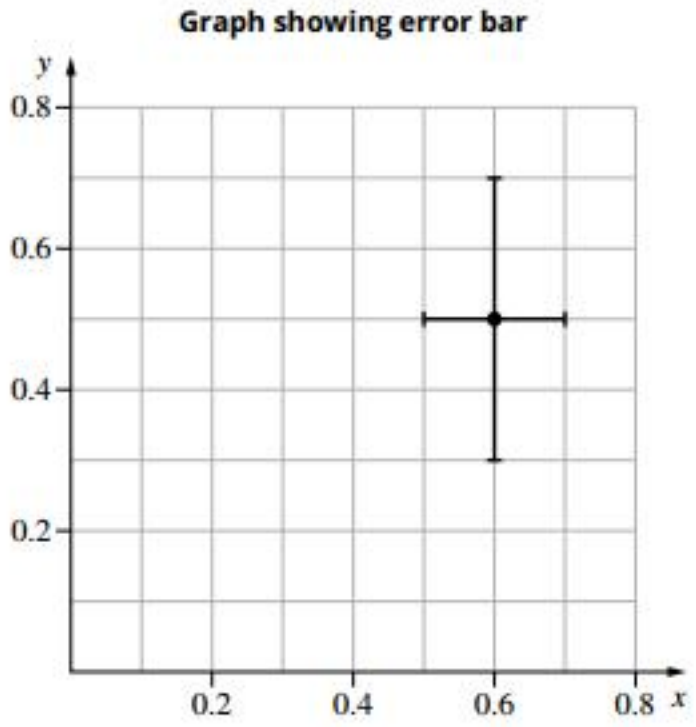


FIGURE 1.6.4 This error bar has an *x* value of 0.6 ± 0.1 and a *y* value of 0.5 ± 0.2. This example is exaggerated for the purpose of illustrating the process of adding error bars.

Error bars are the graphical representation of the uncertainties in the independent and dependent variables.

The line of best fit (trend line), error bars, extrapolation and outliers

The straight line or a curve that best summarises the data is the trend line, also known as line of best fit. This line does not have to pass through every point or the origin (0, 0 point) but does have to be one smooth continuous line or curve (Figure 1.6.5) and *not* a 'join the dots' line (Figure 1.6.2). The line will be drawn by ruler or freehand to best fit the points.

Typically, the line of best fit should pass through as many points as possible, but if this is not possible then it should have as many points above the line as there are below the line (Figure 1.6.5). The trend line may be extrapolated back to the origin (0, 0) or to the *y*-axis (where $x = 0$) but *only* if that makes sense for the data. When making a graph from a table of data, do not plot 0, 0 unless it is given in the table or makes obvious sense for the variables. In Figure 1.6.5 extrapolation back to 0, 0 (the origin) makes sense whereas in Figure 1.6.6 it does not have meaning for these variables, so only the measured data is plotted.

Extrapolation is when a trend or pattern is shown on a graph and you predict beyond the plotted data, by continuing the trend line back or forward from what was measured. This is done to estimate what the result would be if further testing were done.

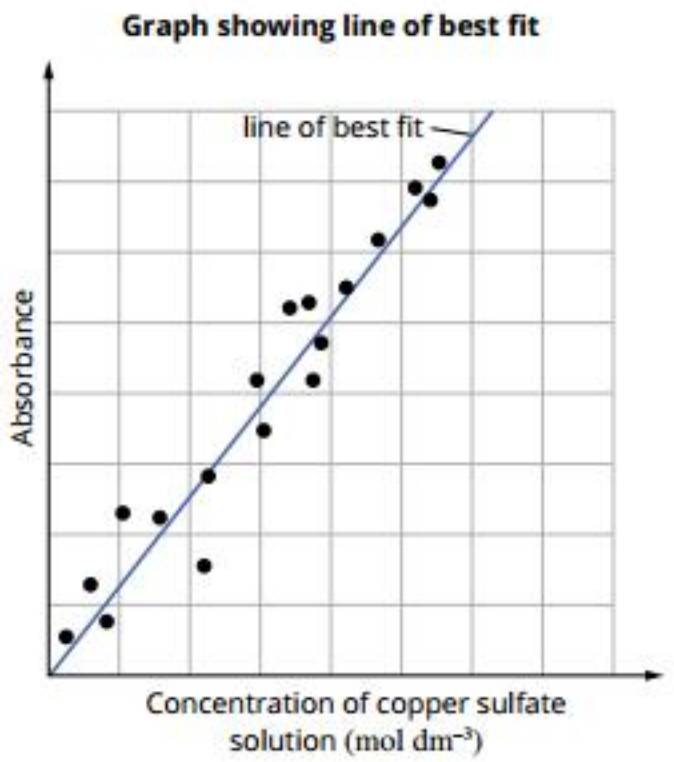


FIGURE 1.6.5 This graph shows that the line of best fit should normally pass through the error bars of each point and be extrapolated so that it intersects the y-axis, if that makes sense for the variables.

The line of best fit is a straight line or curved line that best summarises the general pattern (trend) of the data on the graph.

Error bars/whiskers

When graphically representing data and results, the uncertainty, error or range in data can be shown. When data is collected or measured, the precision or uncertainty is recorded as $\pm n$. This should be shown in the graph. Inserting error bars or whiskers onto the data points on a graph will display a range of n above and below each data point (Figure 1.6.6). This range can be the range in measurements, instrumental precision, uncertainty, error or standard deviation for example, as explained in Module 1.5.

As you will recall from Module 1.5, uncertainty and error are a range in which the true value lies. A graph should indicate this. Choosing which range to indicate on the graph depends on the analysis and interpretation of data trying to be achieved in answering the research question or testing of a hypothesis. Module 1.7 explains various calculations that are useful for analysing and interpreting data and will assist with choosing which range to indicate on a graph to demonstrate reliability or significance of results.

Anomalies and outliers

Sometimes one data point does not fit the trend and may be an error. This is called an **anomaly** or **outlier**. Outliers are often caused by a significant random error when measuring (see Module 1.5). Outliers are determined using statistical calculations (Module 1.7), and they cannot be determined using a trend line. If you have an anomaly or outlier in your results, include it in your graph but ignore it when drawing the trend line of best fit (Figure 1.6.6 and Figure 1.6.3b).

You will learn more about extrapolation and outliers in Module 1.7.

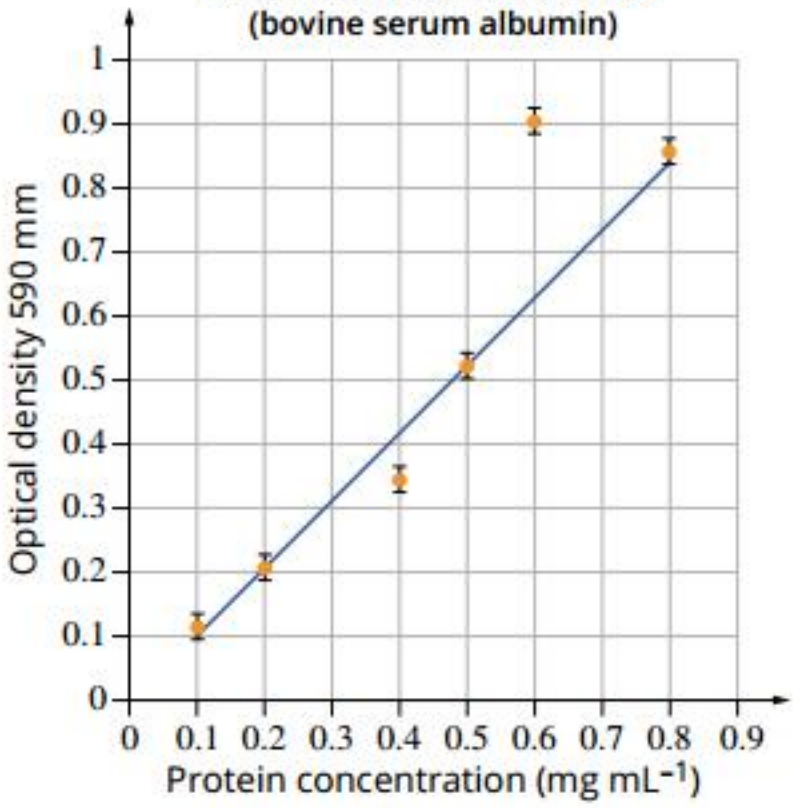


FIGURE 1.6.6 A line graph showing error bars and an outlier (anomaly), which has been ignored when adding the trend line of best fit. The line has not been extrapolated back because there was no data to justify doing that.

Missing data

When you have missing data, leave a gap for it, as shown in Figure 1.6.7. Ensure that the axes are complete, do not skip values and do not join the data points that have missing data points between them.

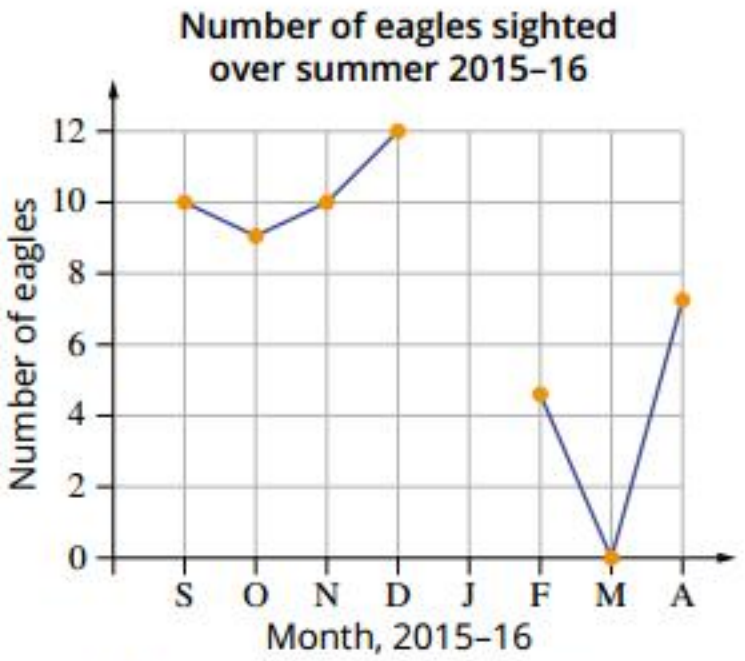


FIGURE 1.6.7 A line graph with missing data indicated by a gap. This accurately represents the measurements taken during the methodology.

Distorting the truth

Poorly constructed graphs can distort the truth. For example, in Figure 1.6.8, you can see two graphs that show the same data—the test results of two groups of students. One group of students did not eat breakfast before doing the test and scored an average of 42 marks out of 50. The other group of students did eat breakfast and scored an average of 48 marks out of 50. One graph distorts the difference in marks between the two groups by using a scale of only 40 to 50 marks on the *y*-axis. It is important to make sure the graphs you create do not distort your data. You should also be wary of distorted data when interpreting graphs in other publications.

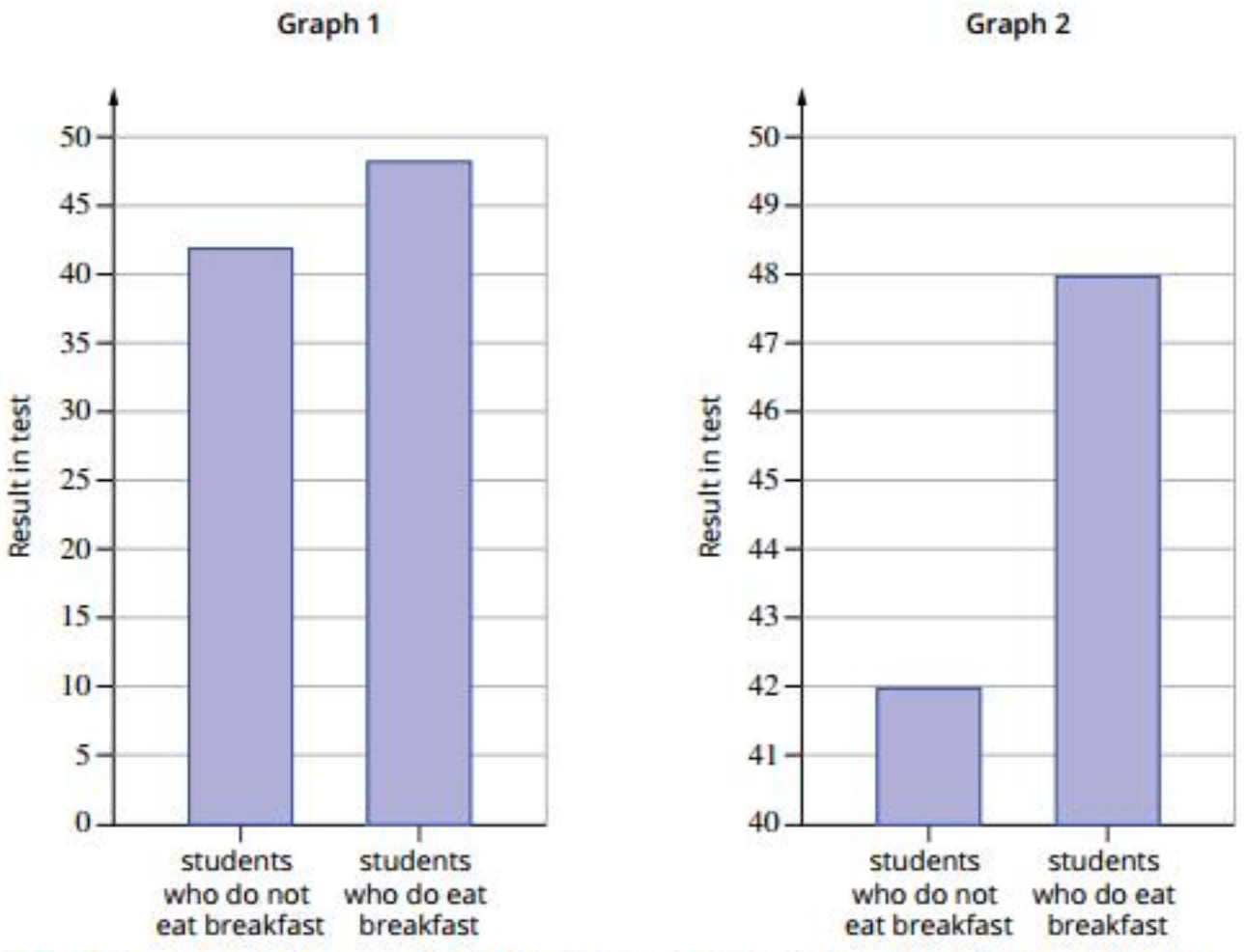


FIGURE 1.6.8 Graph 1 shows the difference between two groups of students out of the total 50 marks on the *y*-axis. Graph 2 shows the difference between the same two groups within only a narrow range of marks on the *y*-axis, which distorts the difference and makes it appear larger than it really is.

These are some of the general rules to follow when making a graph.

- Keep the graph simple, uncluttered and fill most of the grid.
- Use a descriptive title.
- Represent the independent variable on the *x*-axis and the dependent variable on the *y*-axis.
- Start each axis at zero (but do not plot a 0, 0 data point unless one exists).
- Match the length of the axes to the data. This will help fill most of the grid.
- Clearly label axes with both the variable and the unit in which it is measured.
- Use small precise symbols such as circles or crosses for data points and add error bars where appropriate.
- Use different symbols for different datasets e.g. circles for one line and crosses for a second line plotted on the same axes.

TYPES OF GRAPHS SEEN IN BIOLOGY

You will encounter all of these graphs at some stage during your Biology course and develop skills in making and interpreting them.

Scatterplots and line graphs

Scatterplots are commonly used to display data in the form of a graph, and can be used to plot raw or processed data. They are used to show the relationship between two variables when one variable is dependent on the other.

The independent variable, which is set by the experimenter, is always shown on the *x*-axis. The dependent variable, which is the variable measured in the experiment, is always shown on the *y*-axis. The data is plotted on the graph as a series of points. Each point should be drawn in pencil as a small circle or cross. Alternatively, you can use a good computer program to generate your graphs.

A **line graph** is a good way of representing **continuous quantitative data**. In a line graph, the values are plotted as a series of points on the graph. A line can then be drawn from each point to the next, as shown in Figure 1.6.2. This line shows the change in data from one point to the next but does not predict the value of a point between the plotted data.

Alternatively, a single straight or curved line of best fit can be drawn, as shown in Figure 1.6.3 which is a **trend line**. It is used to show the overall trend in the data and can be used to predict values between the data points. A line of best fit usually does not pass through every data point. Its position can be estimated by eye (ensuring an equal number of data points above and below the line when drawn), but mathematical or graphing programs can be used to determine the line of best fit more accurately. This will be covered in more detail in Module 1.7.

Bar and column graphs

Bar and column graphs are used to show categories and **discrete (discontinuous) data**.

- A **column graph** shows the value of the dependent variable by the height of the column; the categories are labelled across the *x*-axis.
- A **bar graph** shows the value of the dependent variable by the length of the horizontal bar; the categories are labelled up the *y*-axis.

Bar and column graphs are commonly used when the independent variable is categorical (qualitative groups e.g. species, colour, soil type) rather than numerical, or when the numerical data is discrete. The bars or columns are always the same width and the same distance apart.

Bar and column graphs are very useful for graphing qualitative and discrete data, such as the water quality data from various locations along a waterway (Figure 1.6.9). Extra columns can be added to the same graph, using colours or shading to compare data. This requires careful planning and plotting of the data (Figure 1.6.10).

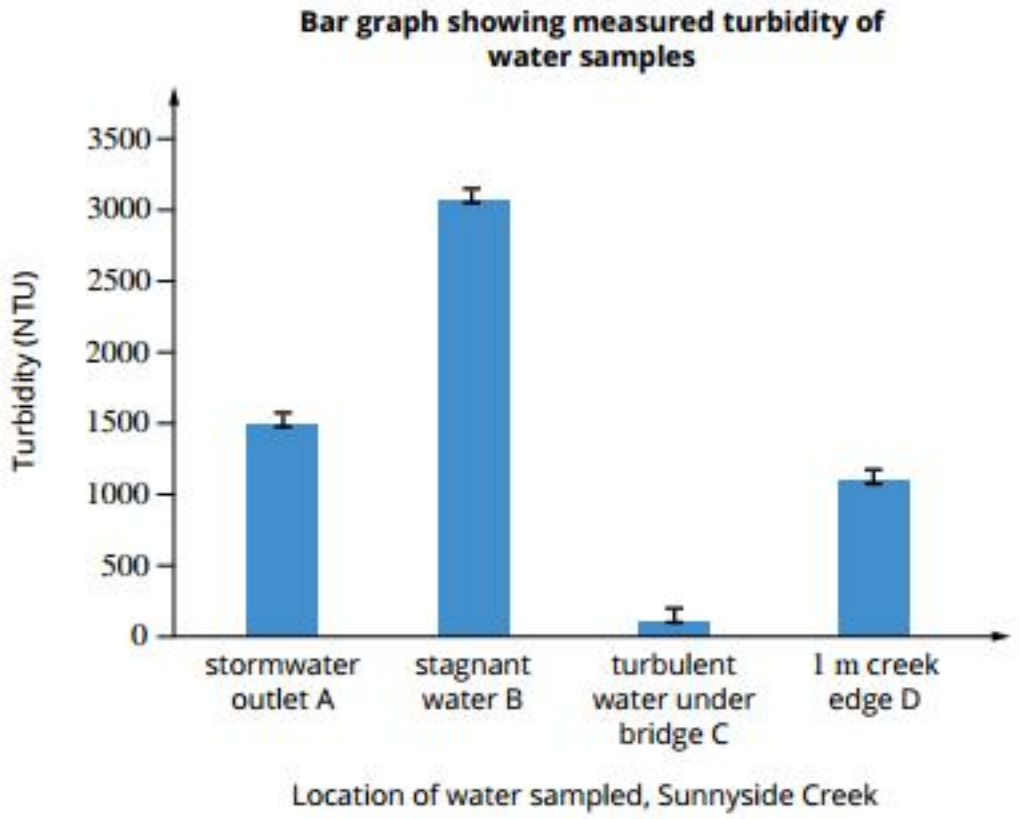


FIGURE 1.6.9 A column graph can be used to compare data with error bars added at the top of each column. This graph shows the measured turbidity of water samples taken from various locations along Sunnyside Creek.

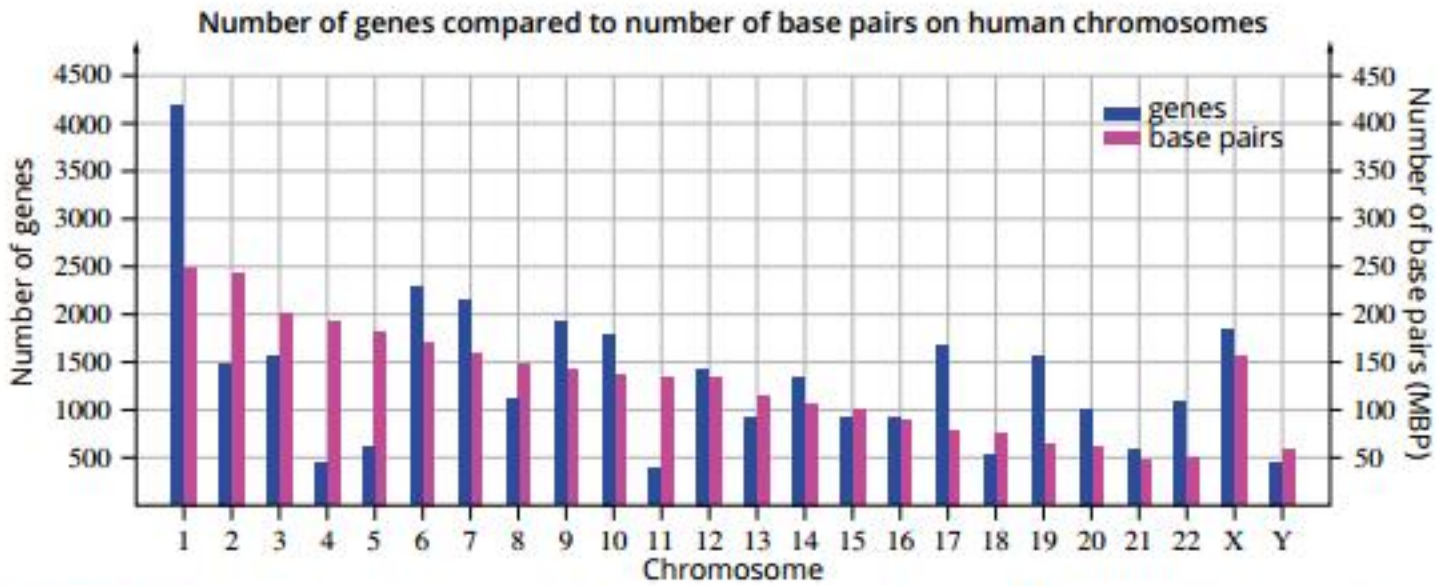


FIGURE 1.6.10 A column graph comparing the number of genes (blue, left axis) and the number of base pairs (pink, right axis) on human chromosomes.

Pie charts

A **pie chart** is a way of presenting qualitative or discrete quantitative data. It shows each category of data as a proportion of the total data. The chart is a circle divided into sections according to the proportions of each category, like slices of a pie (Figure 1.6.11). Each category is coloured or shaded differently to distinguish it from the other categories. It is recommended to use pie charts only when there are a few categories.

A circle is equal to 360°. To draw a pie chart, you must determine how many degrees are needed for each category. This can be done as follows:

1 Add the amounts in each category to find the total.
2 Divide 360° by the total (this will tell you how many degrees of the circle one value is worth).
3 Multiply the answer by the amount in the first category (e.g. the number of students who ate breakfast). Your answer will be in degrees that can then be marked for the first category using a protractor on the circle.
4 Repeat for each category.

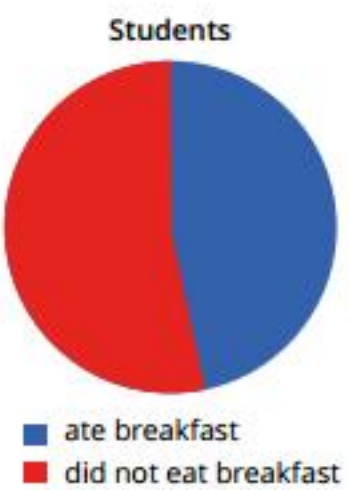


FIGURE 1.6.11 A pie chart presenting data on the breakfast habits of students

1.6 Review

- Tables and graphs are used to represent scientific data.
- In general, tables provide more detailed data than graphs, but it is easier to observe trends and patterns in data with graph format than in table format.
- Tables organise data into rows and columns and can vary in complexity according to the nature of your data.
- Tables should have the following features:
  - a descriptive title
  - column headings (including the units)
  - aligned figures (align the decimal points)
  - the independent variable placed in the left column
  - the dependent variable placed in the right column.
- Graphs are used to display relationships, trends and patterns between two variables with one proposed to be dependent on the other.
- General rules to follow when drawing a graph include to add a title and label both axes with the variable and units.
- The independent variable (IV) goes on the *x*-axis (horizontal) and dependent variable (DV) goes on the *y*-axis (vertical).
- There are several types of graphs. The best one to use will depend on the nature of the data.
- Scatterplots are commonly used to display data in the form of a graph and can be used to plot raw or processed data.
- A trend line (line of best fit) is drawn for the overall trend in the data and can be used to predict values between the data points.
- A line graph is a good way of representing continuous quantitative data.
- Bar and column graphs are used to show categories and discrete (discontinuous) data.
- A pie chart is a way of presenting qualitative or discrete quantitative data for a small number of categories.
- Missing data is represented in graphs by leaving a gap.
- An outlier is a data point that does not fit the trend, and may be caused by an error.

KEY QUESTIONS

Describe

1 State the purpose of a trend line.

2 Clarify what an outlier is.

Apply

3 Identify the relationship between the variables indicated by a sloping linear graph.

4 Explain how the general pattern (trend) of a graph can be represented once the points are plotted.

5 Describe the relationship that exists if both variables increase or both decrease at the same rate.

6 Immunologists have measured the levels of antibodies in blood serum to gather background data on population responses to infection. They collected the following data on the concentration of two different types of antibody, IgG and IgA, from participants ranging in age from 6 months to 20 years (the antibody levels are listed in order of increasing age of subject):

- Age of subject: 6 months, 1, 2, 4, 10, 20 years
- Concentration of IgG (mg/100 mL): 300, 600, 800, 1000, 1500, 1500
- Concentration of IgA (mg/100 mL): 50, 100, 100, 150, 200, 400.

a Prepare a data table.

b Prepare a graph of the data.

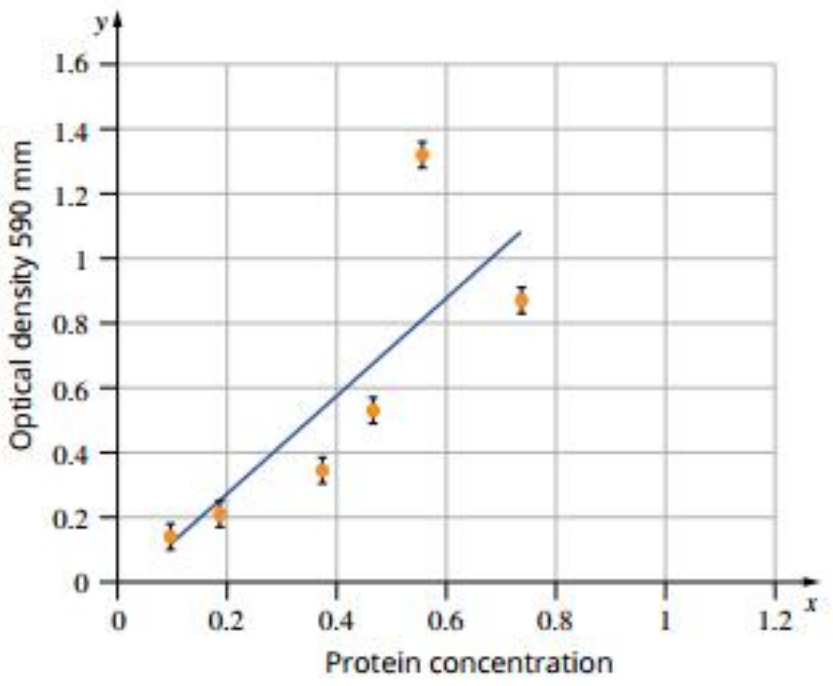
Analyse

7 A student draws a straight-line graph of data, including error bars, from an experiment they have just completed. They find that it is not possible to draw a straight line of best fit through every error bar. Analyse the problem and advise them on what to do.

8 If theory suggests that the *y*-intercept of a line is zero, deduce what this should look like on a linear graph.

9 Assess when it is more appropriate to use a graph showing a line of best fit rather than a ruled line graph joining the line from point to point.

10 Determine at least four ways that the following graph could be improved.



1.7 Data analysis

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- calculate and interpret the range, mean, median and mode of a set of data
- explain the statistical significance of standard deviation
- explain the meaning of regression, linear regression, *r*-values and R^2 values
- understand that various statistical methods are used to analyse datasets, including Student's *t*-test, Pearson's coefficient and Spearman's rank.

One of the difficulties in the biological sciences is that direct measurements, or measurements within a closed system, are challenging to obtain. Therefore, there are various limitations and uncertainties in biology, commonly involving accuracy and precision, as well as errors related to reliability and validity (see Module 1.5). To address this problem, biologists perform statistical analysis to help understand the quality of the measurements and their meaning.

This module will outline a basic understanding of common statistical analysis used in biology and the methods relevant to the QCAA Biology General Senior Syllabus for Units 1 to 4.

STATISTICAL MEASURES

There are a number of common statistical measures that help describe data accurately.

Range

The range is the difference between the highest and lowest values in a dataset. Table 1.7.1 shows the measurements taken for five different plants after treatment with a plant hormone.

TABLE 1.7.1 Plant height in a hormone treatment experiment

Treatment	Height (mm)					Mean (mm)	Range (mm)
	Plant 1	Plant 2	Plant 3	Plant 4	Plant 5		
Hormone-treated	158	378	320	377	363	319.2	378 – 158 = 220
Untreated control	140	135	170	171	193	161.8	193 – 135 = 58

To determine the range for values in Table 1.7.1 you subtract the smallest value from the largest value. Notice how an abnormally large or abnormally small value in the dataset makes the variability appear high.

If one value appears very different, such as plant 1 in the hormone-treated group, the experiment should be repeated more times, or another statistical analysis used to determine whether the unexpected value of 158 mm is an outlier. An outlier is an unexpected value that lies outside (much smaller or larger) most of the other values in a set of data. To determine if a value lies outside most other values, it must be statistically calculated. This illustrates the importance of observation during experiments and recording all details in a journal as the experiment is conducted. The range outlines the total variation in measured results. No other analysis or interpretation can be applied.

Measures of central tendency

Measures of central tendency (also called central location) are single values that allow you to describe the central position in a set of data. Knowing the central or typical value in a dataset can help reveal trends in the data as well as outliers.

The mean, median and mode are all measures of central tendency.

Consider the following dataset: 3, 5, 7, 8, 8, 8, 10.

- The **mean** (or average) is the sum of the values divided by the number of values, which in this case is (3 + 5 + 7 + 8 + 8 + 8 + 10) ÷ 7 = 7.

$$\text{mean} = \frac{\Sigma(x_1 + x_2 + x_3 + \ldots)}{n}$$

where Σ = sum of
$x^1 + x^2 + x^3$ = measurement$_1$ + measurement$_2$ + measurement$_3$
n = total number of measurements

- The **median** is the 'middle' value in an ordered list of values, which in this case is the fourth value, which is 8.
- The **mode** is the value that occurs most often in a list of values, which in this case is 8. This measure is particularly useful for describing qualitative or discrete data.

The appropriate measure of central tendency to use depends on the type of data you are working with (Table 1.7.2).

TABLE 1.7.2 When to use the different measures of central tendency

Type of data	Mean	Median	Mode
Nominal (qualitative)	✓	✗	✗
Ordinal (qualitative)	✓	✓	maybe
Discrete or continuous (quantitative)	✓	✓	✓

Graphically representing the data using central tendency provides a clear and succinct display of results. As there are many measured values and results in raw data, it can be difficult to analyse and interpret. It is much easier and less complicated to analyse and interpret processed data compared to raw data. Raw data includes all the measured results and usually a large number of measurements or observations.

Processed evidence is usually a smaller number of results that represent all the measured raw data; these representative results are calculated using statistical methods such as mean, median and mode. For instance, an analysis can be conducted to compare results using three results (e.g. three means) rather than nine results (e.g. nine individual measurements in the raw data). Interpreting a relationship is also much easier and less complicated when using fewer numbers.

It is important to realise that statistical calculations do not produce the true value, but rather a single value (or smaller number of values) that is meant to represent many.

As can be seen in Table 1.7.3, the two means are processed values of six measurements each. The measured values for heart rate have been averaged using the mean.

STANDARD DEVIATION

Standard deviation (SD or the sigma symbol σ) specifically estimates the spread of data in a population or set of values. It calculates the spread or dispersion of data and its distance from the mean.

The standard deviation calculation assumes that the data being used is from a set in which a normal curve (or normal distribution) exists. A normal distribution of data forms a 'bell-shaped' curve or a normal curve as seen in Figure 1.7.1. A normal curve on a graph has:

- approximately 68.2% of all the measurements (data) within 1 standard deviation (1σ) of the mean; that is, 34.1% above the mean and 34.1% below the mean
- approximately 27.2% of all the data between the first and second standard deviation (between 1σ and 2σ); that is, 13.6% above 1σ and 13.6% below 1σ
- approximately 4.2% of all data between the second and third standard deviation (between 2σ and 3σ); that is, 2.14% above 2σ and 2.14% below 2σ
- the final 0.2% above and below 3σ, 0.13% on either side.

Scientists are careful using the generic word 'average' which refers to any measure of central tendency. In everyday use, when average is mentioned it most often refers to the mean. The mean is a specific type of average, where you add up all the data values and divide by the number of value measurements.

TABLE 1.7.3 Measured resting heart rate (beats per minute) for males and females

Resting heart rate (bpm)	
Female	Male
77	69
78	68
72	71
73	72
76	71
74	68
Mean = 75	Mean = 70

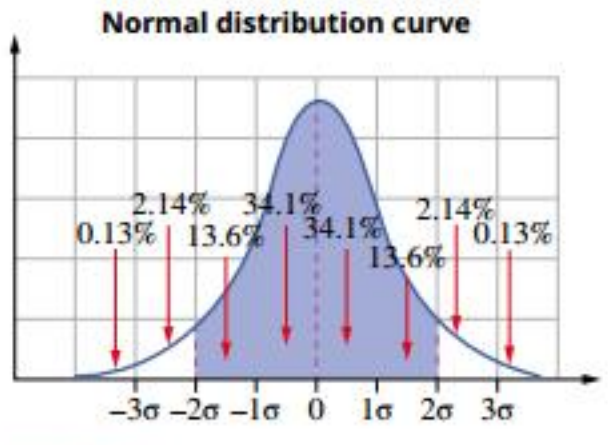


FIGURE 1.7.1 A normal distribution curve showing how far each standard deviation (s) is from the mean (0).

When using standard deviation to analyse data, it is important to understand what has been calculated. If a measured phenomenon is expected to be equivalent from one test to another, then it would be expected that all measurements would be very close to the mean, except for unusual results such as an error. Using SD, you could decide that all accurate measurements should be within two standard deviations (2σ) of the mean. Therefore, any measurement outside (greater than) 2σ from the mean would be considered as an outlier (measurements that have a value outside of the expected calculation). Dealing with a possible outlier when plotting a set of points on a graph was explained in Module 1.6. Using SD will determine if it is a genuine outlier.

If standard deviation was used for a set of data in which variation is expected, it could be used to demonstrate the variation. If the heights of all the female students in a Year 11 Biology class were measured, the results could look like those in Table 1.7.4. A standard deviation could be used to estimate where individuals fit into the assumed normal curve for the height of adult females in Australia.

TABLE 1.7.4 Height of female students in a Year 11 Biology class

Sample	Height (cm)
student 1	164.5
student 2	156.7
student 3	172.2
student 4	180.6
student 5	163.4
student 6	158.8
student 7	153.9
student 8	150.7
student 9	160.4
student 10	156.8
Mean	161.8

Using the formula to calculate standard deviation (SD):

$$SD = \sqrt{\frac{\Sigma(x_1 - \bar{x})^2 + (x_2 - \bar{x})^2 + (x_3 - \bar{x})^2}{n - 1}}$$

where Σ sum of

x_1 = measurement$_1$

$\bar{x}$ = mean

$n - 1$ = number of measurements − 1

The standard deviation for the set of data is ±8.5 cm. Student 4 at 180.6 cm is more than 2s away from the mean (Figure 1.7.2). Therefore, it can be said that this student is estimated to be in the top 3% of the adult female population for height. This is not an outlier because variation is expected, so standard deviation was used to understand the variance of female students in the class.

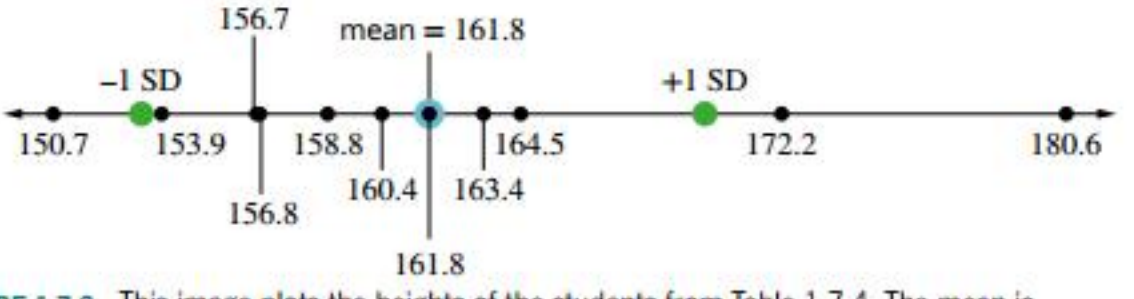


FIGURE 1.7.2 This image plots the heights of the students from Table 1.7.4. The mean is represented by the blue dot; one standard deviation from the mean on either side (±8.5 cm) is represented by the two green dots

Worked example 1.7.1

CALCULATING THE MEAN AND STANDARD DEVIATION

The heights of a group of students were measured and are listed below. Calculate the mean and standard deviation for student height:
160 cm, 158 cm, 123 cm, 145 cm

Thinking	Working
Write the formula for the mean.	$\bar{x} = \frac{\sum x_1 + x_2 + x_3 \ldots}{n}$
Insert the values and perform the calculation.	$\bar{x} = \frac{160 + 158 + 123 + 145}{4}$ $= 146.5 \text{ cm}$
Write down the formula for standard deviation.	$SD = \sqrt{\frac{\sum (x_1 - \bar{x})^2 + (x_2 - \bar{x})^2 + (x_n - \bar{x})^2 +}{n-1}}$
Insert the values and perform the calculation.	$SD = \sqrt{\frac{\Sigma(160-146.5)^2 + (153-146.5)^2 + (123-146.5)^2 + (145-146.5)^2}{4-1}}$ $= \sqrt{\frac{\Sigma 182.25 + 132.25 + 552.25 + 2.25}{3}}$ $= 17 \text{ cm}$

➤ Try yourself 1.7.1

CALCULATING THE MEAN AND STANDARD DEVIATION

The following data was collected for the number of offspring that mice produced. Calculate the mean litter size and standard deviation.

Litter sizes: 2, 14, 6, 8, 9, 11, 5, 6

REGRESSION ANALYSIS

Regression is the term used to state that the independent variable causes the result in the dependent variable during an experiment.

There are numerous statistical calculations that estimate the causal relationship between variables.

- Some calculations indicate how directly an independent variable causes the dependent variable result. This attempts to show the strength of the relationship.
- Other statistical calculations for regression consider the variation in results and attempt to predict the effect the independent variable has on the dependent.

In biology, due to the open system nature of many experiments, not all relationships are direct or straightforward cause-and-effect. Often, there are many variables that influence the dependent variable and it is helpful to try and predict how much of an effect the independent variable has on the dependent variable while other variables are also influential. The mathematical techniques that follow are required in the QCAA syllabus for Biology.

Pearson's correlation coefficient

Linear regression is a calculation that estimates a direct linear relationship between the independent and dependent variables. It assumes a direct causal relationship. When used, a straight line (trend line of best fit) can be drawn on a graph using all the data points (results) and an *r* value is displayed.

In statistics, r values represent correlations between two numerical variables. The value of r is always between +1 and −1, and can be anywhere in between. For example, the r value could be −0.92, −0.78, −0.13, 0, 0.24, 0.81 etc. A positive r value means that the independent variable causes an increase in the dependent value (Figure 1.7.3). A negative r value means that the independent variable causes a decrease in the dependent value (Figure 1.7.3). The r values of −1 and 1 mean the relationship is perfectly correlated and therefore 100% causal, r values of −0.91 or 0.91 means the correlation between the variables is strong at 91%, and values of −0.13 or 0.13 would be due to a weak relationship that is 13% correlated. A value of 0 means there is no relationship. Remember that the negative and positive values refer to the type of relationship (positive refers to an increasing relationship and negative to indicate a decreasing relationship).

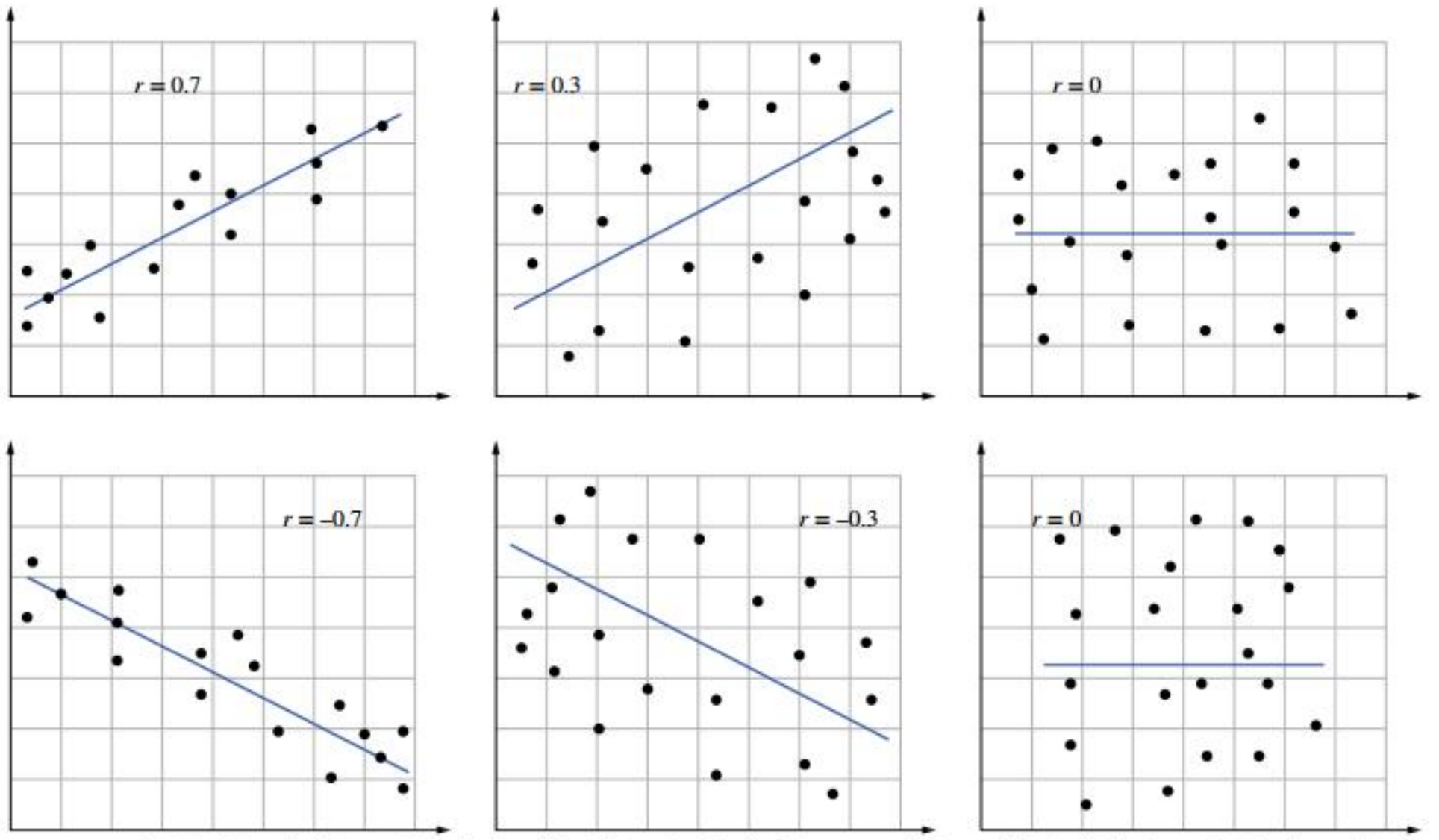


FIGURE 1.7.3 Scatterplot graphs demonstrating the line of best fit produced by the Pearson correlation coefficient with their associated r value written on each grid. Notice the difference between an r of 7 and 0.

The **Pearson correlation coefficient** calculates the strength of the relationship; hence, the r value is an indicator of how strongly the dependent variable is related to the independent variable. If the relationship is strong and no errors occurred during experimentation, then it would be expected that the experimental data (results) would be plotted near the trend line (the x and y intercepts) with an r value above 0.8 or between −0.8 and −1.0. Different disciplines of science have different interpretations of the r value. Some interpret values of 0.5 to 1.0 or −0.5 to −1.0 to be strong, while others require values above 0.75, or from −0.75 to −1.0, to be considered strong. The closer the value is to 0, the weaker the relationship is (Figure 1.7.3).

Along with establishing how strong the direct relationship is, you can also make a forecast and extrapolate information. **Extrapolation** is when a trend or pattern is shown on a graph and you predict beyond what the graph is showing, by continuing the trend or pattern beyond what was measured. This is done so that you can estimate what the result would be if further testing were done. You will recall that extrapolation on graphs was introduced in Module 1.6.

The Pearson correlation coefficient can use either processed or raw data in its calculation. Raw data is best because the result of the calculation is a more accurate representation of the relationship. However, it is important to note that the calculation assumes that outliers have been removed. This would have to be achieved using a different statistical calculation first, such as standard deviation (explained above).

Examples of using the Pearson correlation coefficient to analyse data are shown in Tables 1.7.5 and 1.7.6. One experiment tested the effect of exercise intensity on heart rate while the other tested the effect of temperature above 45°C on the rate of photosynthesis.

The r value for the Pearson correlation coefficient was calculated using the formula:

$$r = \frac{n(\Sigma xy) - (\Sigma x)(\Sigma y)}{\sqrt{[n\Sigma x^2 - (\Sigma x)^2][n\Sigma y^2 - (\Sigma y)^2]}}$$

where n = the number of samples (measurements)

x = dependent variable value on the x axis

y = independent variable value on the y axis

Completing statistical calculations is time consuming and due to multiple inputs can easily result in errors. Most statistical calculations can be done easily using software programs such as Excel.

Coefficient of determination (R^2)

The **coefficient of determination** (R^2) can calculate a value that indicates the predictability between the variables based on how much the results vary. The R^2 produces a value that is an estimate for how well you can predict the dependent variable based on the independent variable, considering the variations in the results. In the examples in Figure 1.7.4, the data from using technique 1 does not exactly follow the linear trend line. Therefore, as the concentration changes (the independent variable), the absorbance (dependent variable) does not always change consistently. Sometimes it changes by 0.05, sometimes by 0.02. The coefficient of determination (R^2) is a calculation that considers the variation in the results to estimate the predictability.

For example, a laboratory has used two different analytical techniques to determine the concentration of a performance-enhancing drug in a blood sample from an athlete suspected of cheating. The calibration curves for each technique and the R^2 value for each are shown in Figure 1.7.4.

Based on the lines of best fit and R^2 values, the second method was chosen to analyse the athlete's blood sample.

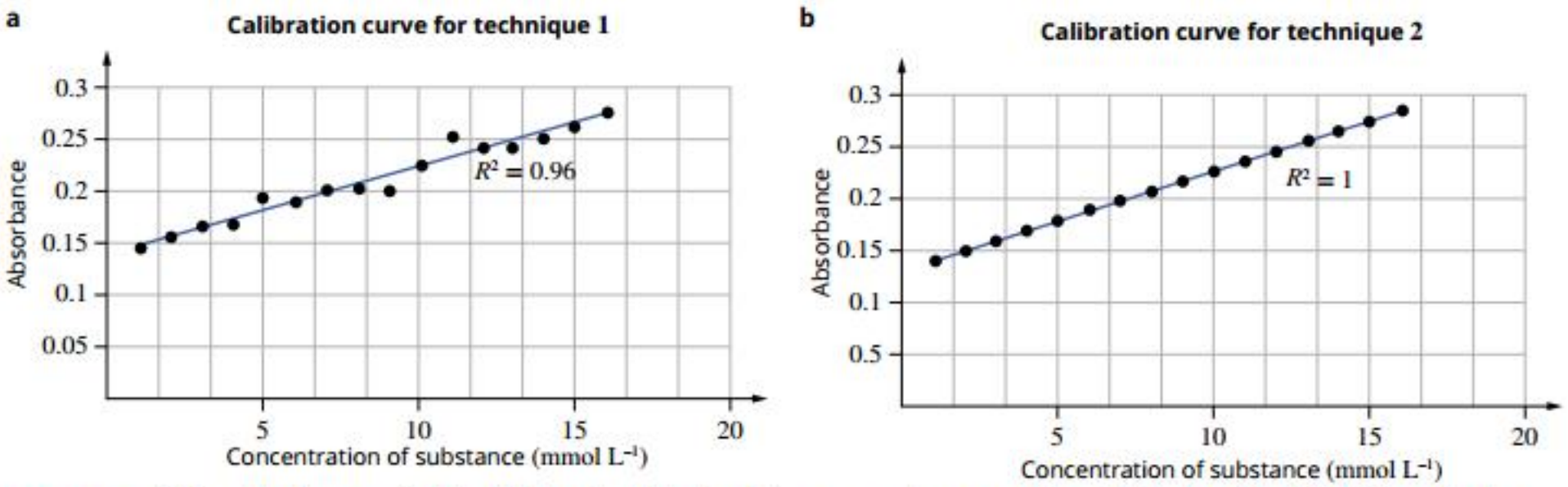


FIGURE 1.7.4 (a) The calibration curve for blood testing using technique 1: linear regression was used to establish a line of best fit. (b) The calibration curve for technique 2: linear regression was used to establish a line of best fit

Even though both techniques measured the same samples, the data is different. As can be seen in the graphs, the linear trend line is very similar with a similar slope; however, technique 1 has an R^2 value of 0.96 and technique 2 is 1.00.

The coefficient of determination produces a value between 0 and 1. If the value is 0, it is unable to predict the change in the dependent variable from the independent variable due to so much variation in the results. If the value is 1, then it can predict the change in the dependent variable 100% of the time without error. Any other number between 0 and 1 is considered a percentage, for example 0.76 means that the coefficient of determination can predict 76% of the dependent variable results due to the independent variable. It is interpreted as 76% of the variation in the dependent variable can be explained by the independent variable.

In Figure 1.7.4b, the absorbance of the sample was perfectly correlated without error with the concentration of the species. This suggests that the results from technique 2, including error, **bias** and instrumental precision, are highly reliable.

Figure 1.7.4a shows that the absorbance of the sample was not precisely correlated with the concentration. The R^2 value for technique 1 is lower than in technique 2, meaning the results are less predictable due to more variation. Technique 1 was less precise compared to technique 2; therefore, technique 2 is the preferred technique to determine the concentration in the actual sample.

In experiments, there could be a number of reasons for the R^2 value to not be above 90% or close to 100%, and not all reasons should be considered due to poor results or deficiencies in the experimentation. A scientist may be trying to establish that no relationship exists and therefore would hope to produce a low R^2 value. Or they may be trying to establish if any relationship exists but it is not known how strong or direct the relationship may be and therefore an R^2 value between 0.50 and 0.90 would be valuable.

The formula to calculate the R^2 value (coefficient of determination) is:

$$R^2 = \left(\frac{1}{N} = \frac{\Sigma[(x_i - \bar{x}) \times (y_i - \bar{y})]}{(\sigma_x \times \sigma_y)^2}\right)$$

where N = number of measurements (values)

x_i = x value (independent variable)

$\bar{x}$ = mean for x value (independent variable)

y_i = y value (dependent variable)

$\bar{y}$ = mean for y value (dependent variable)

σ_x = standard deviation of x

σ_y = standard deviation of y

Computer software and apps have formulas to conduct statistical calculations, making it easier to perform such analysis. Also, fewer errors are made when calculations are conducted using software, provided the initial data is entered correctly.

Pearson correlation coefficient versus coefficient of determination

Both statistical calculations produce a trend line of best fit and both analyse only a linear relationship. Table 1.7.5 summarises the two statistical analytical tools.

TABLE 1.7.5 Summary of the two statistical analytical tools

	Pearson correlation coefficient	Coefficient of determination
Calculates	How closely variables are related (correlated).	How predictable the change in the dependent variable is due to the independent variable.
Result	r value = –1.00 to 1.00	R^2 value = 0 to 1.00
Meaning	The closeness of the relationship is given a value and whether the independent variable positively or negatively influences the dependent variable.	Provides a value that indicates the variation that always occurs in experimentation and influences the results and the validity or reliability of the data.
Interpretation	The strength of the relationship (correlation) is due to a causal relationship either directly or indirectly.	The higher the value, the less variation in the data, therefore the more reliable or valid the data.

An analysis of Figure 1.7.5 using the r and R^2 values could be interpreted as: the r value of 0.97 shows a strong relationship between the length of time in days and the length of a leaf (cm). The relationship between time and leaf length is causal. The R^2 value of 0.93 is high, with 93% of the increase in leaf length being predictable from the change in time. The biological process for growth in leaf length is impacted by many variables, such as photosynthesis, cellular respiration, water and nutrient access and supply. However, the data analysis reveals that the predictability of leaf length (cm) due to time (days) is high. In this experiment, very few variables influenced the results more so than time. This could mean that the biological processes are closely related to time. Or, as there is little variation in the data from the linear trend line, the results are highly reliable.

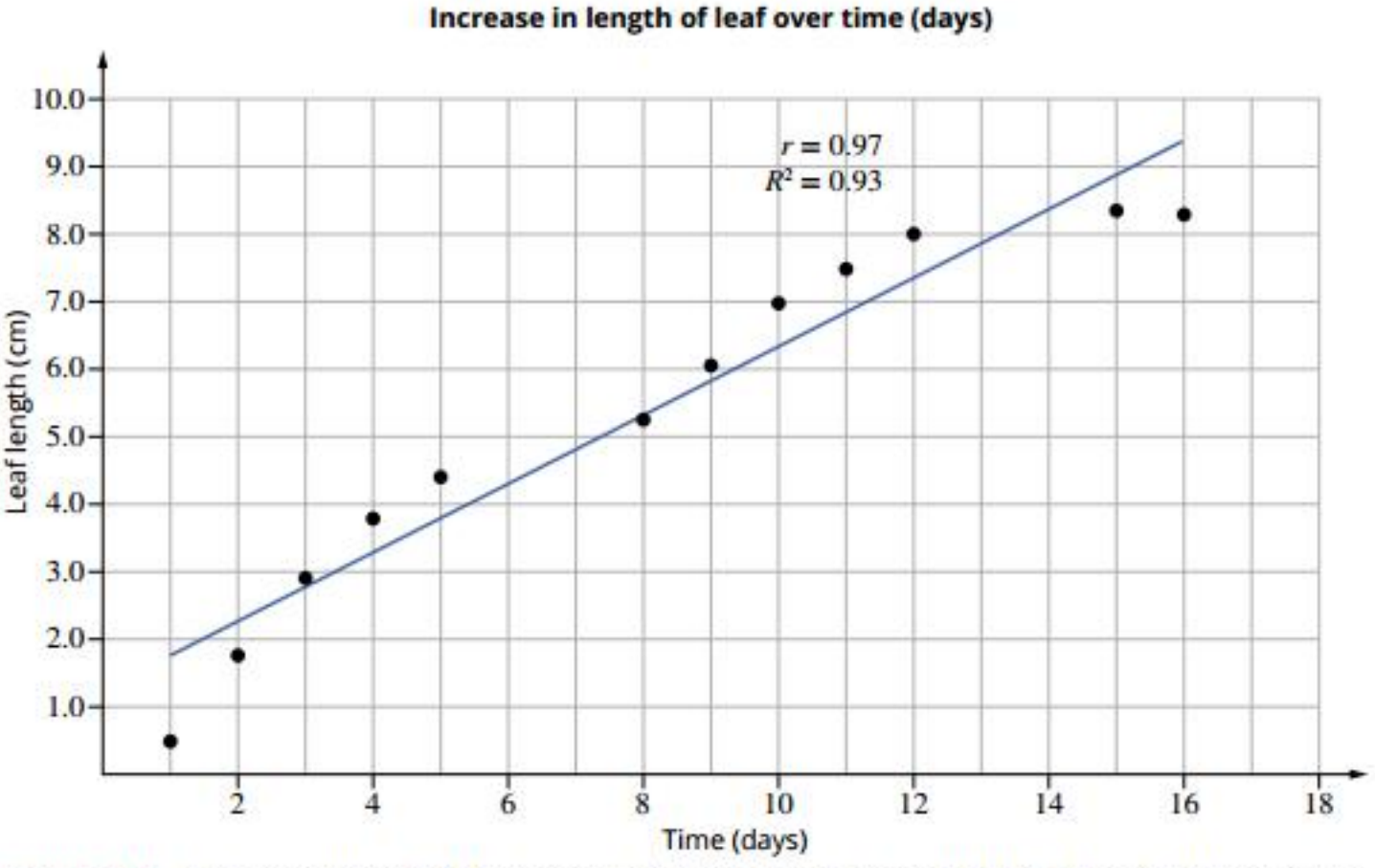


FIGURE 1.7.5 Scatter plot showing the length of a leaf over a number of days with the r value of 0.97 and R^2 value of 0.93 displayed

Spearman's rank correlation

Spearman's rank correlation is another calculation that determines a trend line. However, it can be used for non-linear regression relationships, as seen in Figure 1.7.6. This calculation estimates the strength and direction of a non-linear relationship between two variables in a single direction. The results of ±–1.0 to ±1.0 are interpreted the same as the Pearson correlation coefficient (Figure 1.7.7).

Spearman's rank correlation works by calculating Pearson's correlation on the ranked values of the related datasets. Ranking (from low to high) is obtained by assigning rank numbers of 1 to the lowest value, 2 to the next lowest and so on. As already noted, computer software and apps have formulas for statistical calculations, making it easier, faster and more accurate to perform such analysis, provided the initial data is entered correctly.

The formula for Spearman's rank correlation is:

$$r_s = 1 - \frac{6\sum d_i^2}{n(n^2 - 1)}$$

Where:

r_s is the coefficient

n is the number of data points

d_i is the difference in the ranks of the two coordinates

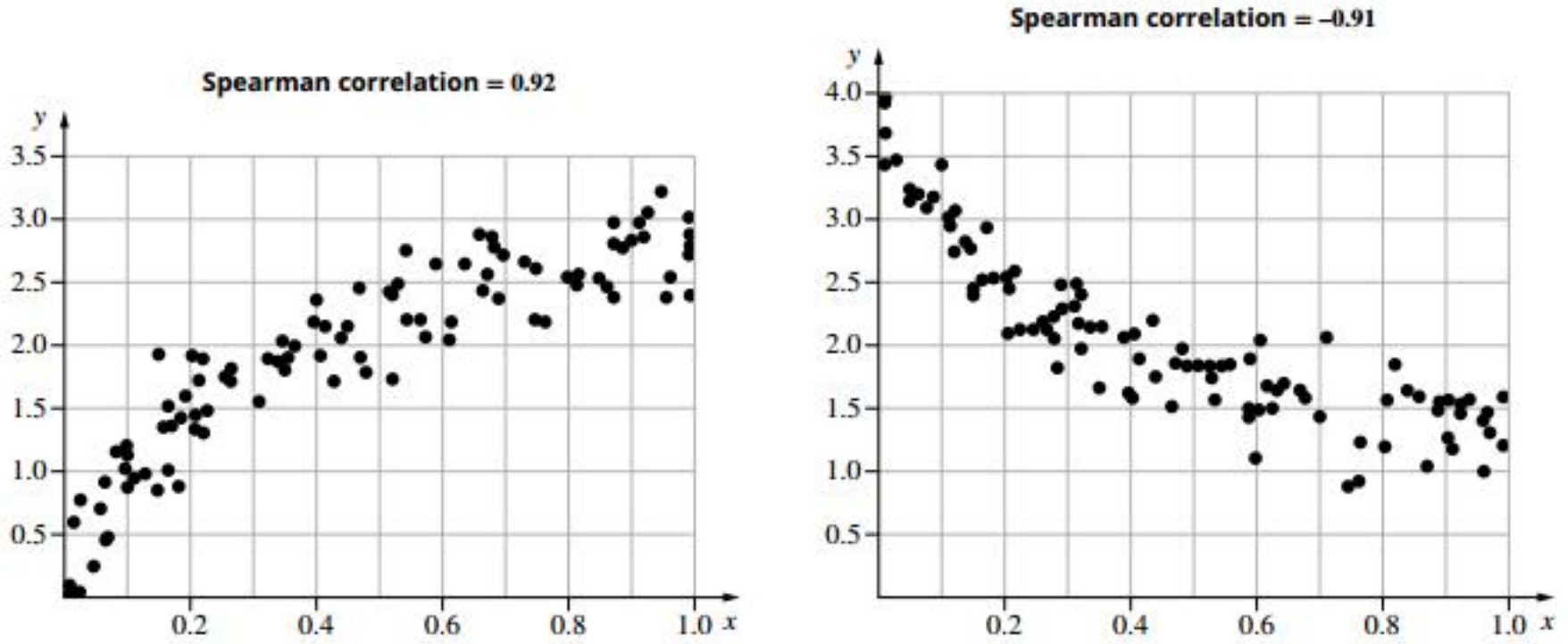


FIGURE 1.7.6 The data in these graphs follows a non-linear pattern, and using the Spearman correlation, the graph on the left has a 0.92 (positive) correlation, meaning as the independent x variable increases, so does the dependent y (in a non-linear fashion). The graph on the right shows a negative correlation, meaning as x increases, y decreases in a non-linear pattern.

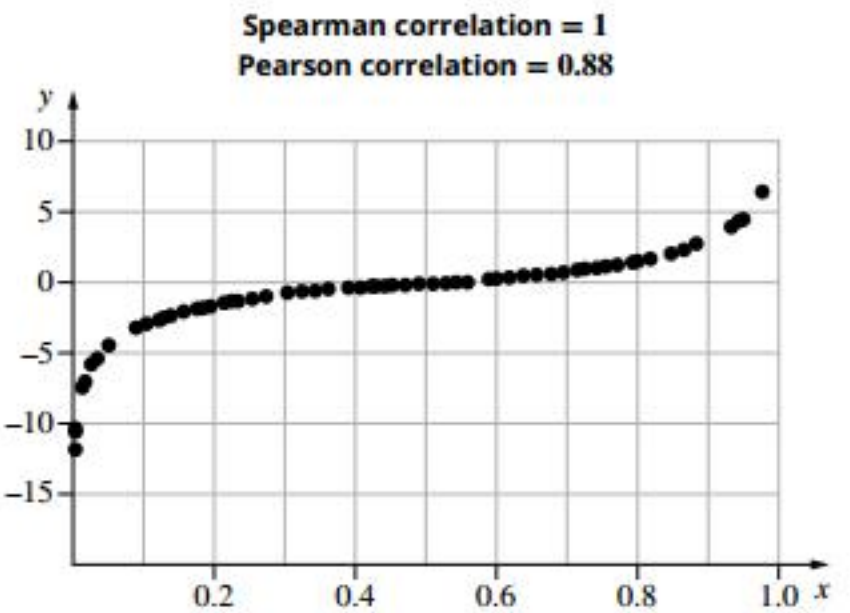


FIGURE 1.7.7 An example of how the Spearman rank correlation explains the pattern in the data more precisely than the Pearson correlation coefficient. Therefore, the relationship is non-linear

STUDENT'S *t*-TEST

The **Student's *t*-test** (also known as the *t*-test) compares two means to determine if there is a difference between them. More specifically, the *t*-test is used for a null hypothesis (a hypothesis that no change or difference will be seen in the results) and estimates the probability that the difference between the means is due to natural variation or by chance. As the null hypothesis assumes no change between a set of results when the independent variable is changed, the *t*-test assumes the null hypothesis is true and that any variation between the sets is due to natural variation, and not the independent variable.

The calculation for the Student's *t*-test produces a *t*-value (also known as *p* for probability) between 0 and 1.00 and is a percentage. For example, a *t*-value of 0.04 is 4% and a *t*-value of 0.82 is 82%. A *t*-value below 0.05 means that there is less than 5% chance (p = probability) that the difference between the means of two sets of data is due to natural variation. You would therefore reject the null hypothesis; you would reject that there is no difference between the two means (therefore, the sets of results are significantly different).

A *t*-value of 0.43 (may also be reported as $p = 0.43$) means that there is a 43% probability that natural variation or chance caused the difference between the means. The calculation of *t*-values above 0.05 do not provide a strong enough estimation to reject the null hypothesis, so no significant difference between the means would be the conclusion.

One of the unique features of the Student's *t*-test is that it can be used for **small sample sizes**, although it is limited to being used for samples (datasets or sample groups) that are related.

There are two types of Student's *t*-tests, unpaired and paired (Table 1.7.6). An unpaired Student's *t*-test compares the means and datasets from two different samples. A paired Student's *t*-test compares the means and datasets of the same sample tested twice, before and after changing the independent variables.

TABLE 1.7.6 Examples of unpaired and paired Student's *t*-tests

Unpaired Student's *t*-test	Paired Student's *t*-test
comparing the mean heights of the high school swim team and the athletics team	comparing the mean height of the 14-year-old boys in the athletics team one year with their mean height a year later
comparing the mean chloroplast counts of a sample of *Elodea* from one pond compared to a sample from another pond	comparing the mean chloroplast count of a sample of *Elodea* from fresh water with the same sample after being treated with salt for 2 weeks
comparing the mean white blood cell counts of a sample of people exposed to the flu in Brisbane and a sample from Townsville	comparing the mean white blood cell count of a sample population in Brisbane, and then the same sample population 2 months later

It is best to use software such as Excel, IBM SPSS Statistics® or your graphics calculator to calculate the *t*-value.

Worked example 1.7.2

USING THE STUDENT'S *t*-TEST: LIGHT AND SHADE

An experiment was conducted to test if sunlight has an effect on the number of chloroplasts found in the cells of *Elodea* plants. A null hypothesis was developed, that there would be no difference in the number of chloroplasts between plants grown in natural light and those grown in shade. Calculate the *t*-value to determine if the null hypothesis is supported or not.

Thinking

Record the raw data from the experiment and place it into Excel.

Working

Plant	No. of chloroplasts per cell	
	Natural daylight	Shade
A	13	11
B	15	13
C	16	15
D	23	14
E	18	12
F	19	16

Thinking

Calculate the mean, using Excel 'insert function'.

Working

Plant	No. of chloroplasts per cell	
	Natural daylight	Shade
A	13	11
B	15	13
C	16	15
D	23	14
E	18	12
F	19	16
Mean	17	14

Thinking

Summarise the results.

Working

The mean number of chloroplasts for Elodea plants grown in natural light is 17, while the mean for plants grown in the shade is 14. There is a difference between the means but it is not known if it is significant. Both groups include variation: Elodea grown in natural light have a range of 10, while those grown in shade have a range of 5.

Thinking

Choose the *t*-test in Excel 'insert formula'. Select sample 1 (plant A results), then sample 2 (plant B results) and so on. Choose 2 tail, and then 2 again for the type of *t*-test.

Working

Plant	No. of chloroplasts per cell	
	Natural daylight	Shade
A	13	11
B	15	13
C	16	15
D	23	14
E	18	12
F	19	16
Mean	17	14
t-value	0.04	

Summarise the results of the *t*-test	There is a 4% chance that the difference between the means and datasets is due to natural variation. Therefore, the difference in the mean number of chloroplasts is significant. The null hypothesis is not supported, and therefore rejected.

➤ Try yourself 1.7.2

USING THE STUDENT'S *t*-TEST: COLOUR OF LIGHT

An experiment was conducted to test if the colour of light has an effect on the number of chloroplasts found in the cells of *Elodea* plants. One group of plants was grown in sunlight while the other group was grown under red light.

Sunlight results: 14, 18, 19, 21, 24, 19

Red light results: 13, 14, 15, 15, 17, 17

A null hypothesis was developed, that there would be no difference in the number of chloroplasts between plants grown in natural sunlight and those grown in red light. Calculate the *t*-value to determine if the null hypothesis is supported or not.

Worked example 1.7.3

USING THE STUDENT'S *t*-TEST: SALT

An experiment was conducted to test if placing *Elodea* in salt water for two days would affect the number of chloroplasts found in the cells of *Elodea* plants. A null hypothesis was developed, that there would be no difference in the number of chloroplasts between the plants in fresh water and the plants in salt water. Calculate the *t*-value to determine if the null hypothesis is supported or not.

Thinking	Working
Record the raw data from the experiment and place it into Excel.	
Calculate the mean, using Excel 'insert function'.	

Plant	No. of chloroplasts per cell	
	Fresh water	Salt water
A	13	12
B	15	13
C	16	16
D	23	17
E	18	18
F	19	16

Plant	No. of chloroplasts per cell	
	Fresh water	Salt water
A	13	12
B	15	13
C	16	16
D	23	17
E	18	18
F	19	16
Mean	17	15

<table>
<tr><td>Summarise the results.</td><td>The mean number of chloroplasts for Elodea plants grown in fresh water is 17, while the mean for plants placed in salt water is 15. There is a difference in the means but it is not known if it is significant. Both groups include variation, Elodea grown in fresh water have a range of 10, while those grown in shade have a range of 6.</td></tr>
<tr><td>Choose the t-test in the Excel ‘insert formula’. Select sample 1 (plant A results), then sample 2 (plant B results). Choose 2 tail, and then 1 for the type of t-test.</td><td>
<table>
<tr><th rowspan="2">Plant</th><th colspan="2">No. of chloroplasts per cell</th></tr>
<tr><th>Fresh water</th><th>Salt water</th></tr>
<tr><td>A</td><td>13</td><td>12</td></tr>
<tr><td>B</td><td>15</td><td>13</td></tr>
<tr><td>C</td><td>16</td><td>16</td></tr>
<tr><td>D</td><td>23</td><td>17</td></tr>
<tr><td>E</td><td>18</td><td>18</td></tr>
<tr><td>F</td><td>19</td><td>16</td></tr>
<tr><td>Mean</td><td>17</td><td>15</td></tr>
<tr><td>t-value</td><td>0.08</td><td></td></tr>
</table>
</td></tr>
<tr><td>Summarise the results of the t-test.</td><td>There is an 8% chance that the difference in the means is due to natural variation. Even though a notable difference exists between the two tests, it cannot be interpreted as significant as it is above 5% (0.05) and may not be due to the exposure to salt water. The hypothesis is supported.</td></tr>
</table>

➤ Try yourself 1.7.3

USING THE STUDENT’S *t*-TEST: BLUE LIGHT

An experiment was conducted to test if placing Elodea under blue light for two days after being grown in natural light would affect the number of chloroplasts found in the cells of Elodea plants.

Blue night results: 22, 19, 21, 21, 24, 16

Natural light results: 23, 18, 20, 21, 24, 19

A null hypothesis was developed, that there would be no difference in the number of chloroplasts after exposure to blue light. Calculate the t-value to determine if the null hypothesis is supported or not.

1.7 Review

SUMMARY

- The range is the difference between the highest and lowest values in a dataset.
- The mean, $\bar{x}$, is the arithmetic average of a dataset. It is calculated using the formula:

$$\bar{x} = \frac{\sum x_1 + x_2 + x_3 \ldots}{n}$$

  where Σ = sum of $x_1 + x_2 + x_3$ = measurement$_1$ + measurement$_2$ + measurement$_3$, n = total number of measurements
- The median is the 'middle' value in an ordered list of values.
- The mode is the value that occurs most often in a dataset. The mode is particularly useful for describing qualitative or discrete data.
- Standard deviation is a measure of the spread or dispersion of data from the mean.
- Regression is the term used to state that the independent variable causes the result in the dependent variable during an experiment.
- Linear regression is a calculation that estimates a direct linear relationship between the independent and dependent variables.
- *r* values (Pearson's correlation coefficients) indicate the degree of correlation between variables.
- *r* values are between –1 and 1.
- A positive r value means that a positive relationship exists between the variables, that is the independent variable causes an increase in the dependent value.
- A negative *r* value means that the independent variable causes a decrease in the dependent value.
- The closer the r value is to 1 or –1, the stronger the correlation between variables.
  - *r* values of -1 and 1 mean the relationship is 100% correlated
  - *r* values of -0.91 or 0.91 mean the correlation between the variables is strong (91%)
  - *r* values of -0.13 or 0.13 are due to a weak relationship (13%)
  - An *r* value of 0 means there is no relationship.
- The coefficient of determination estimates how well you can predict the dependent variable based on the change in the independent variable.
- The coefficient of determination produces an R^2 value between 0 and 1.
- The closer R^2 is to 0 the less predictable the dependent result is based on the independent value.
- The closer the R^2 is to 1 the more predictable the dependent result is based on the independent value.
- The Spearman's rank correlation determines a non-linear trend line and the strength of non-linear relationships, in a single direction.
- The Spearman's rank correlation produces a value equivalent to the Pearson correlation coefficient and is interpreted in the same way. It is based on ranking two related sets of data values.
- The Student's *t*-test assumes that any variation between the sets is natural and is not due to the independent variable.
- Student's *t*-test values are interpreted as a percentage chance (also known as a *p* value). For example, 0.42 is a 42% chance that the variation is natural, caused by chance not by changes in the independent variable.
- Student's *t*-test values less than 0.05 are significant and are caused by the independent variable.

1.7 Review *continued*

KEY QUESTIONS

Describe

1 Define the terms 'mean', 'median' and 'mode'.

Apply

2 Describe what the standard deviation informs scientists about in a dataset.

3 Describe what the Student's *t*-test informs scientists about in a dataset.

Analyse

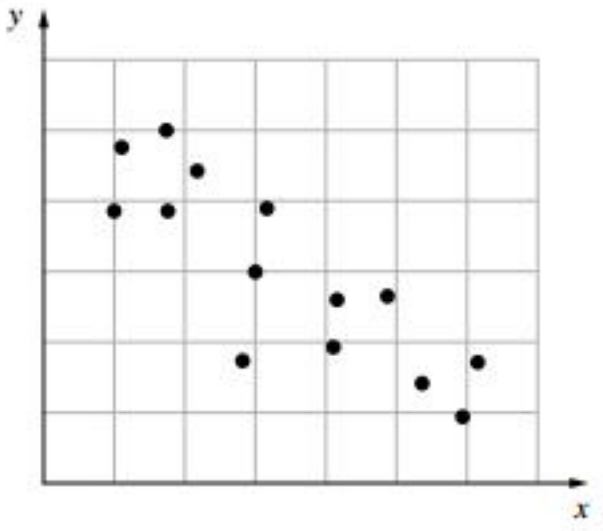
4 For the dataset {21, 28, 19, 19, 25, 24, 20} calculate the:

- **a** range
- **b** median
- **c** mode
- **d** mean
- **e** standard deviation

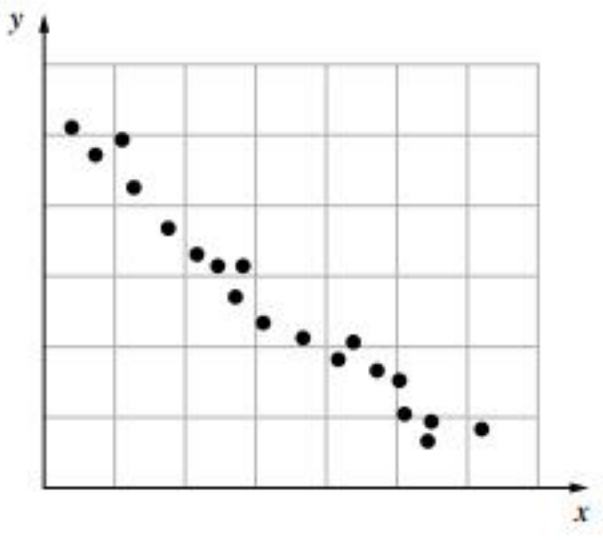
5 Identify which of the statistical measurements of mean, mode and median is generally most affected by an outlier.

6 Select the appropriate *r* value from the list below for each of the following graphs a to d.
r values: –0.90, –0.50, 0.00, 0.50, 1.00

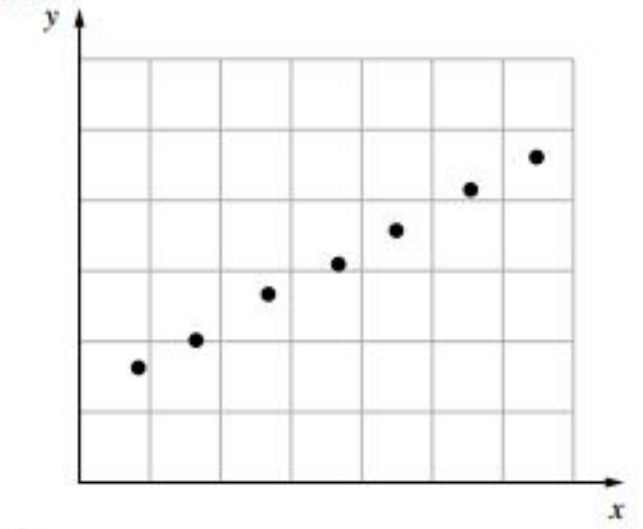
(a)



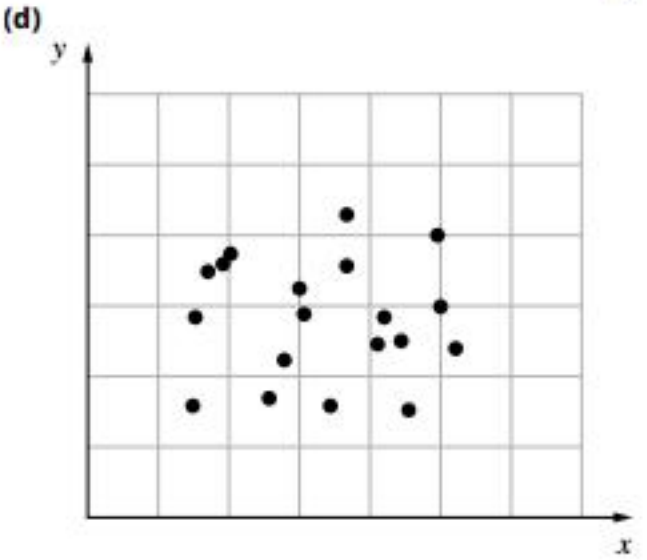
(b)



(c)



(d)



1.8 Scientific writing

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- use scientific language and representations
- use a scientific journal for experiments
- reference sources and avoid plagiarism
- use and reference AI in research and writing
- communicate findings in a multimodal presentation.

The QCAA requires students to complete a student experiment as part of Unit 3 Biology and a research investigation in Unit 4 Biology. In preparation for Units 3 and 4, teachers may choose to assign similar assessment tasks in Units 1 and 2. Both tasks require written or multimodal presentations. Preparing and presenting scientific reports and investigations involves research and note-taking skills, use of scientific genre (writing style), language and representations, and referencing knowledge to acknowledge sources used.

Scientists follow strict genre conventions when undertaking and communicating their work. They are described in this module. Refer back to this module as you complete the student experiment (Part B, Modules 1.9–1.13) and the research investigation (Part C, Modules 1.14–1.18).

SCIENTIFIC WRITING AND REPRESENTATIONS

Scientific reports must be written using scientific language and representations – they should be relevant, fluent and concise. To meet the word count or time requirements, only use the words required to communicate your information and avoid repetition (Figure 1.8.1). Having a plan and using a journal (logbook) also helps to achieve fluent and concise writing. Use the syllabus objectives, specifications and the instrument specific marking guide (ISMG) to plan an outline of each section of your written task. Part B and Part C will support you with writing reports specific to the assessment tasks.

Table 1.8.1 demonstrates communication of the same concept in a few different ways. Being fluent and concise will significantly influence the word count of the report and the quality of the writing.

TABLE 1.8.1 Communicating concepts in various ways

Concept	Communication	Commentary of communication
The basic structure of a cell	A cell is made up of a nucleus and other parts.	This is fluent and concise but lacks scientific terms or language to convey in-depth understanding.
	Cells are tiny little objects made up of numerous parts that form the basis of living organisms. Cells are made up of smaller parts and cytoplasm. The nucleus is one smaller part that is the control centre of the cell and contains genetic material.	This is fluent but not concise. There is repetition e.g. 'tiny' and 'little'—these terms have the same meaning; also 'made up' and 'parts/part' are stated twice or more, and 'living' is redundant when used with organism. It uses words without using scientific terms to provide detail and understanding e.g. 'parts' instead of organelle. This may be useful language in the journal when first learning about a topic. Continued use would distract from learning the correct terms and it should not be used in a scientific report.
	Cells are the basic structural and functional units of living things. They are composed of a membrane around cytoplasm (cytosol with specialised smaller organelles embedded in it). Each cell contains a nucleus with DNA molecules which carry the genetic information for that organism.	This is fluent and concise. Repetition is avoided. The sentence structure flows and has close links for connected terms (e.g. 'cytosol' with 'organelles'). Much in-depth understanding is communicated through use of scientific terms – membrane, cytoplasm *and* cytosol, organelle, DNA. These terms enable relevant information to be conveyed in fewer words *and* demonstrate advanced, accurate knowledge, such as the subtle difference between cytoplasm and cytosol.

Reports are written using paragraphs, with each paragraph explaining only one idea. The first sentence (topic sentence) of the paragraph introduces the topic. Following sentences provide the details of the idea. The final sentence concludes the idea and may also link to the idea of the next paragraph. Each sentence should flow onto the next, slowly building the details of the explanation – similar to how this book is written.

Reports should be written using:

- past tense—as the experiment was conducted in the past, so the report should be in the past tense
- third person, passive voice and impersonal verbs—science uses this language convention (Table 1.8.2)
- scientific language—the terms used are specific to concepts, models and theories. Use of scientific language allows more details, knowledge and understanding to be communicated in fewer words and more accurately.
- information from secondary sources which is written in your own words
- objective, unbiased language—avoid subjective and emotional or persuasive writing (Table 1.8.3)
- concise language—avoid padding with irrelevant words and unnecessary repetition to express ideas. Use shorter sentences that are less verbose (wordy) (Table 1.8.4)
- headings and subheadings, and sometimes dot points, can be used to present an accomplished science report.

TABLE 1.8.2 Examples of first-person and third-person narrative

First person	Third person
I put 50g of marble chips in a conical flask and then added 10mL of 2M hydrochloric acid.	*First, 50g of marble chips was weighed into the conical flask and then 10mL of 2M hydrochloric acid was added.*
After I observed the reaction, I found that ...	*After the reaction was completed, the results showed ...*
My colleagues and I found ...	*Researchers found ...*

TABLE 1.8.3 Persuasive writing versus scientific writing styles

Persuasive writing examples	Scientific writing equivalent examples
Biased and subjective language: *The results are extremely bad/exciting/wrong because ...* *This is terrible because ...* *This produced a disgusting odour.* *Health crisis!*	Unbiased and objective language: *The results showed that ...* *These results imply that ...* *This produced a pungent odour.* *Health issue*
Exaggeration: *The object weighed a colossal amount, like an elephant.*	Non-emotive language: *The object weighed 256kg.*
Use of everyday or colloquial language: *The bacteria passed away.* *The results don't ...* *The researchers had a sneaking suspicion that ...*	Use of formal language: *The bacteria died.* *The results do not ...* *The researchers predicted/hypothesised/theorised, inferred that ...*

TABLE 1.8.4 Examples of verbose versus concise writing

Verbose writing	Concise writing
Due to the fact that ...	*Because ...*
Smith undertook an investigation into ...	*Smith investigated ...*
It is possible that the cause could be ...	*The cause may be ...*
end result ...	*result ...*
In the event that ...	*If ...*
shorter in length ...	*shorter ...*

To produce a fluent report, the scientific language must be used without error so that the reader understands the meaning of the information easily. Refer to the syllabus and your textbook including the glossary for the correct scientific terminology. Being fluent and concise will significantly influence the word count and the quality of writing. Refer to the syllabus ISMG to identify the cognitive verbs used in the descriptors to produce the appropriate response. For example, you will need to write more extensively if you are asked to 'discuss', 'describe' or 'explain' compared to 'identify', 'name' or 'recognise'.

Scientific representations

Scientific models, equations and diagrams are an efficient way of presenting complex data and explaining biological concepts. Flow charts/schematics, graphs, tables and photographs are also used (Figure 1.8.1). Ensure you include:

- a descriptive title
- labels, captions or descriptions, including units of measurement (if appropriate)
- numbering; for example, Figure 1, Figure 2 etc. or Table 1, Table 2 etc.
- a source if the work is not your own or is adapted from work that is not your own.

SCIENCE JOURNAL

There is a lot of information to record during the student experiment or research investigation so it would be best to keep a **scientific journal (logbook)**. Entries in your journal should include the following features:

- the date of entry
- ideas, observations, proposals, questions
- background information you researched for the task
- new terms and definitions
- refinements
- results, data, evidence, variables
- a numbering system for diagrams, tables and graphs
- explanations, ideas, observations (including diagrams)
- limitations, conclusions, further investigation.

A journal holds a record of all information used during the process of the scientific method (Figure 1.8.2). You will not know what is vital information until your experiment is complete and summarised into the scientific report. Data and graphs may be saved digitally, so reproduce a sketch in your journal that you can annotate with your thoughts. Use a numbering or coding system to keep track of tables and graphs, and cross reference these to your journal sketches. You may choose to prepare a page or two each for the four key sections of the task (e.g. for IA2: Forming, Finding, Analysing, Interpreting and Evaluating) or use proposed report headings (e.g. Rationale, Research Question etc.).

Researched and recorded information in your journal will be used for the rationale; to justify the modifications to the methodology; to provide a reason for collecting data; to inform risk, ethical and environmental management; and to collate

and process the data. It will also inform the interpretation of the evidence (results) and conclusion. Remember, parts of the rationale (planning) and the data collection are completed with your group and shared—the rest of the task is individual and so is your journal. A journal is not an assessed part of the task but will likely be reviewed by your teacher as a progress check and to confirm the work is yours. Depending on the size of the journal, it may be copied and attached in the Appendix to your report.

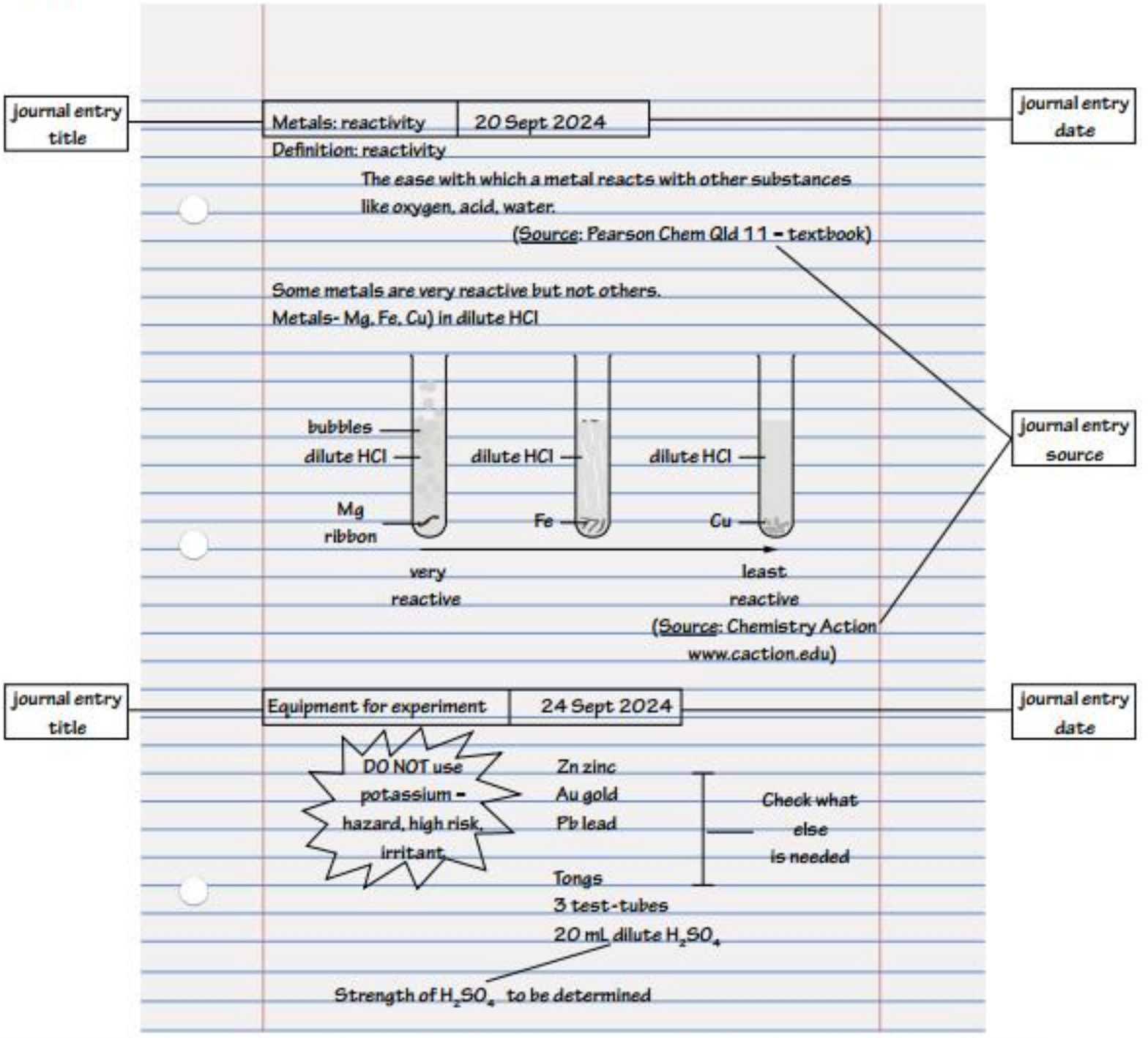


FIGURE 1.8.1 A journal entry with the journal entry date, titles and apparatus used in preparation for a chemistry experiment. A biology journal should use a similar record-keeping process because all areas of science have a common approach for reporting investigations. There will be numerous journal entries on any given date, and they should have their own titles. Also, the same titles will appear on different entry dates as research continues on specific topics.

Journal entries can take different formats. Sometimes one form of recording information provides more details than others. Note taking is not limited to words. Figure 1.8.2 presents the same information in different ways.

a

Protein is synthesised using coded information from DNA. The DNA code is replicated into an RNA strand which is transported to the ribosome (which is at the endoplasmic reticulum). Here the ribosome uses the coded information to build a protein.

b

DNA is copied into RNA (coded information)

↓

RNA transported to Endoplasmic recticulum and Ribosome

↓

Ribosome uses coded information to build protein

c

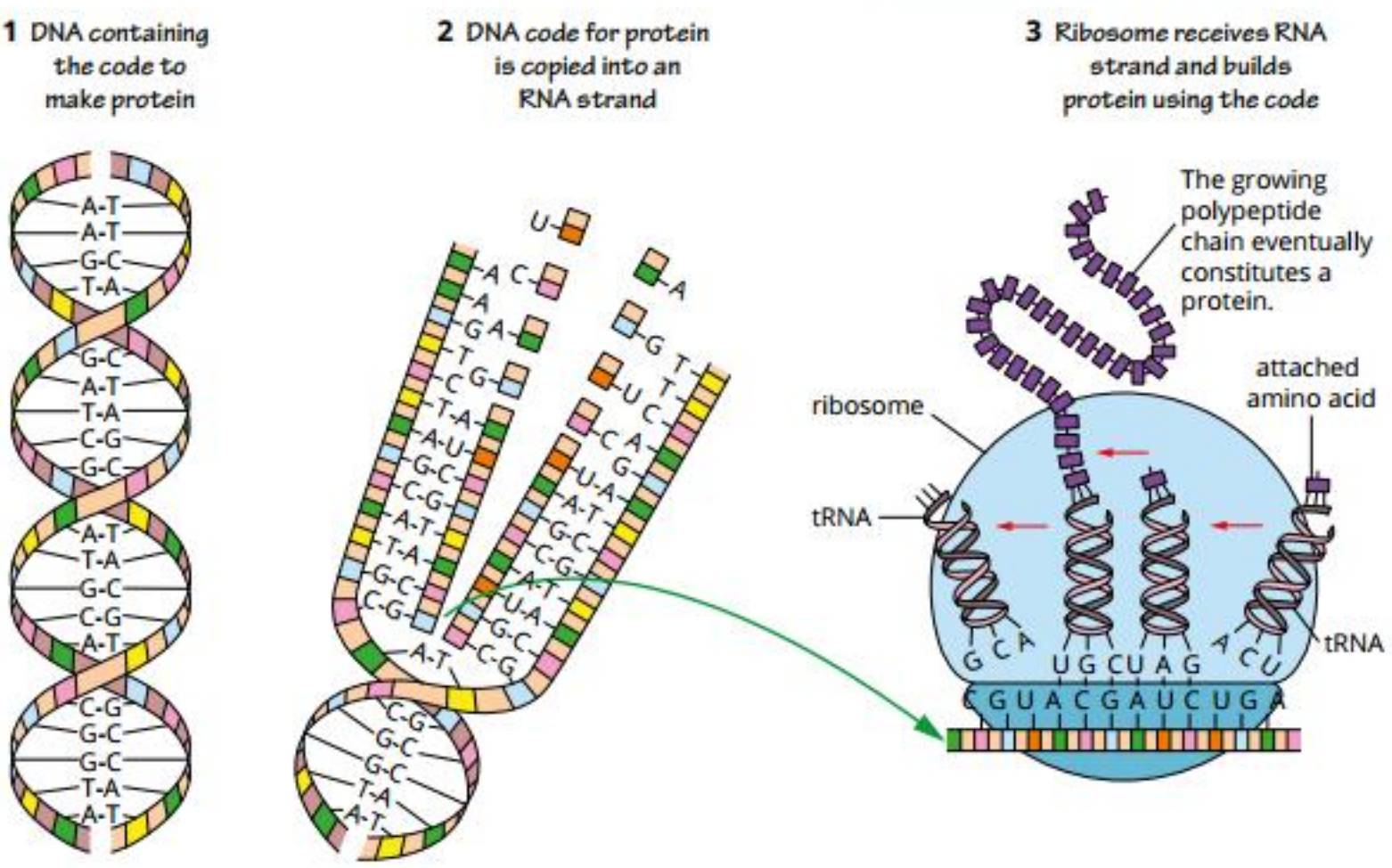
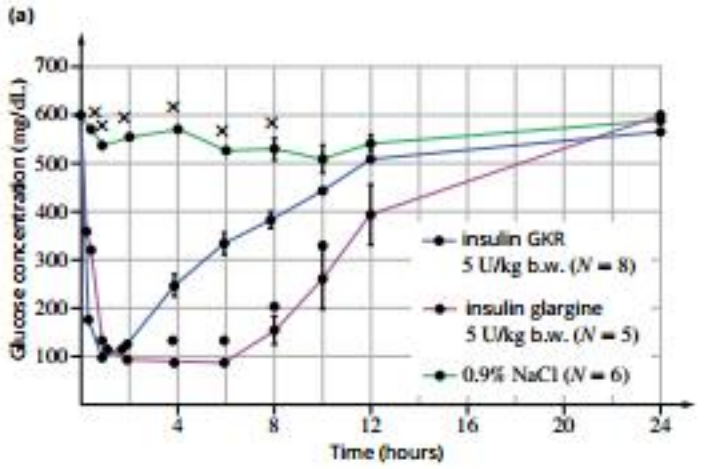


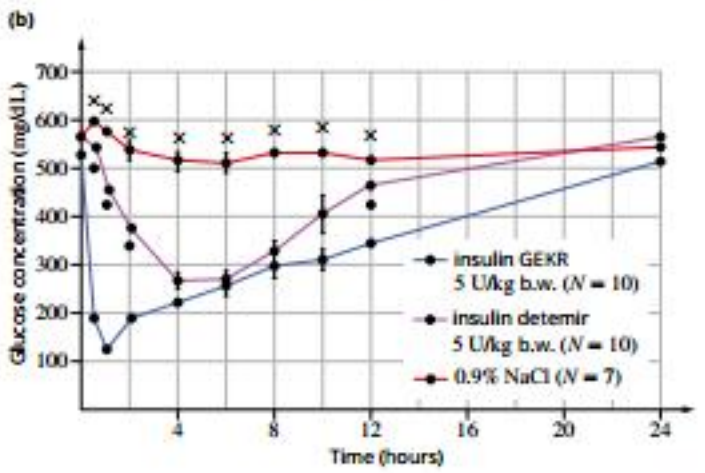
FIGURE 1.8.2 Three ways of representing the same information in a journal. (a) a written summary on protein synthesis, (b) a flow chart showing protein synthesis as a simple schematic, (c) a model diagram explaining protein synthesis.

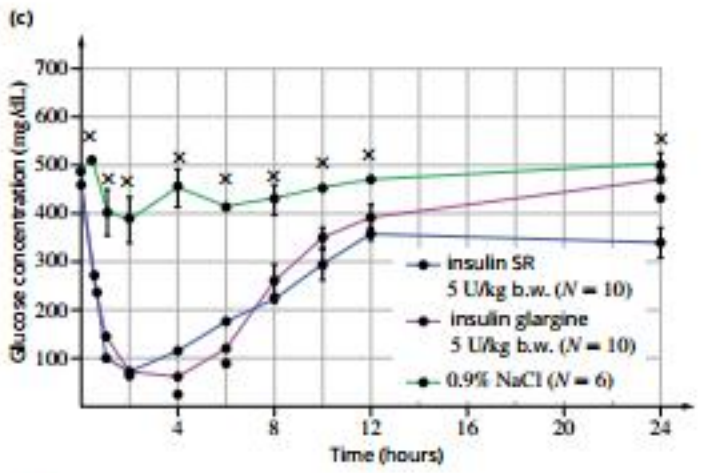
When recording results, data or evidence in your journal, be critical and interpret the data *as it is collected*. In this way, errors in the method are identified quickly or sample size can be adjusted, if needed. If a given group member has the recording task, it is vital that the dataset is passed to all group members before leaving the laboratory. Add this information directly into your journal and annotate with your own comments. Figure 1.8.3 shows an example of research evidence and how it is interpreted in a journal. Notes in your journal can also include:

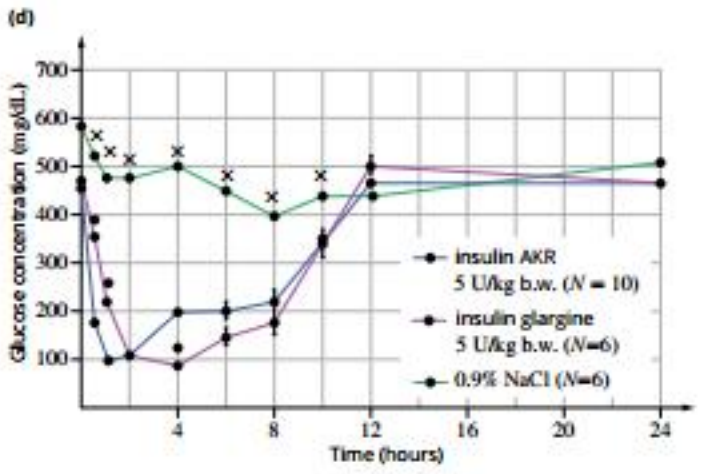
- specific details and your interpretation of significant values
- the trend or pattern and a statement of whether this is the expected one
- the comparison or difference between one set of values and another
- the statistics used to establish the significance and the author's interpretation.

When making journal entries for research data, statistics and methods (e.g. R^2 value, confidence interval, standard deviations, Student's *t*-test, Pearson's coefficient (r) or Spearman's rank), it is important that you express the information in your own words. You will most likely have to conduct further research to understand the statistics, their meaning, and the author's interpretation of the results as you will come across new statistical calculations.









6/7/17

Data analysis

Fig 3 a-d Glycaemia profiles personal interpretation:

Each graph shows how much glucose is in the blood after being injected with a substance (over time). Each graph compares the amount of glucose removed from the blood by a control insulin molecule (already manufactured by companies for diabetics), a new insulin molecule (genetically engineered) and salt.

Graph a)

- Green line salt. Did not remove much glucose from the blood when injected.
- Blue line = insulin GKR 5 (genetically engineered). Removed a lot of glucose from the blood, then returned to normal reasonably quickly over 12min.
- Purple line = insulin glargine (typical manufactured molecule). Removed a lot of glucose from the blood, then returned slowly to normal by 20min.
- Crosses and black circles above data is significant according to $p < 0.5$

What does $p < 0.05$ mean:

Is a statistical calculation that estimates the probability of something occuring within natural variation (assuming a null hypothesis). Here, it is the probability that no difference will exist (except natural variation) between the salt and insulin results and also the GKR5 and glargine results. The crosses and black circles mean the probability of natural variation causing the results in less than 5%.

These results mean that there is more than a 95% chance that these results are due to the independent variable (the different type of insulin). (source: http://www.statsdirect.com/help/basics/p_values.htm)

FIGURE 1.8.3 The interpretation of original primary evidence from a research journal article: (a) primary evidence displayed in graphs with statistics; and (b) student's interpretation in their journal. The *p* value is probability calculated using a Student's *t*-test

REFERENCING SOURCES

When gathering scientific evidence for a scientific inquiry such as Student Experiment and Research Investigation, source it from reputable publications such as:

- your textbook and practical manual. This should be your first source of reliable information
- scientific journals and magazines: research papers and scientific reviews. Common commercial science magazines that you might find in your school library, in print form or as an online subscription, include The Helix, Cosmos, Scientific American and New Scientist. Scientific journals are peer-reviewed which gives them credibility
- podcasts and websites. Electronic sources include online scientific organisations such as CSIRO, NCBI, AIMS, Bioscience (Oxford Academic) and other university publications and press. Many government and privately funded science organisations publish material, along with not-for-profit scientific organisations. Websites may direct you to magazines and scientific journals, podcasts, blogs and videos.

In the syllabus, you are asked to 'discuss and evaluate the validity and reliability of evidence" (Science inquiry skills). When selecting and using sources of information and evidence, follow these guidelines.

1 Confirm that it is relevant.
   - **a** Check that it contains information that is specifically related to the inquiry.
   - **b** Check that the evidence and information are pertinent to the variables of the inquiry.

2 Assess its reliability.
   - **a** Is it current/recent information?
   - **b** Is it up-to-date in its understanding of relationships?
   - **c** Is the evidence in agreement with other sources?
   - **d** Check credibility and bias—consider who the author is, their qualifications, expertise and affiliations. A scientific paper or an article by a reputable journalist will include this information.
   - **e** Evaluate the methodology, including which variables were controlled or measured.

It is not necessary that each of your sources fulfill all of these requirements. Each source will have strong and weak points. For example, we can be confident that information from a peer-reviewed scientific journal has credibility and is unbiased because the review process is undertaken by experts in the field. However, it may have been produced 10 years ago and so scientific information and relationships may not be up to date. Such a source could be used together with a less credible but more up to date one.

Podcasts or science magazines for school students may contain a wide array of interesting real-world examples but not explain the scientific background in sufficient detail. They may have been illustrated and written so as to attract attention but with less scientific detail, and therefore require more in-depth research from other sources. An informed discussion of these types of issues is part of the evaluating section of the Research Investigation. Some resources are available by paid subscription, so use advice from your school librarian about free resources or join your local library.

All quotations, documents, publications and ideas used in your investigation need to be acknowledged in the reference list in order to avoid plagiarism and to ensure creators are properly credited for their work. When using sources to explain scientific concepts or review data, ideas must be written in your own words (see plagiarism, below). Using multiple sources and cross-checking between them will help you evaluate and meet the syllabus requirements. References and acknowledgements show that you have read widely, give credibility to your study and allow the reader to locate information sources should they wish to check or study it further.

Plagiarism

Plagiarism is using other people's work without acknowledging them as the author or creator. To avoid plagiarism, include a reference every time you report the work of others; for example, at the end of a sentence or following a diagram. This is called an in-text citation. If you use a direct quotation from a source, enclose it in quotation marks. This will ensure you give credit to the original author and it will enable the reader to find the original source.

Information taken from a source should be rewritten in your own words. It can be difficult to reword information in your own words. A useful strategy is to read the information, understand it, then, without looking back at the source, take notes in your journal. Notes that are detailed and have full explanations will be useful when you refer to them later to write the report.

TABLE 1.8.5 Examples of paraphrasing or rewording information from a source

Source information:
'It can be expected that the days will be warm and the nights will be cool over the next three days due to the high pressure system moving across the region.'

Strategy to change source information	Reworded information
The order of information can be swapped: the object (the warm days and cool nights) with the reason (high pressure system).	Due to the high pressure system moving across the region, the days will be warm and the nights will be cool.
Some words can be changed such as adverbs, nouns, verbs, adjectives.	It has been predicted that during the daytime it will be warm while the evenings will be cool for about three days, as a high pressure system travels over the region.
Information can be elaborated to explicitly outline information.	The weather forecast predicts that over the next 72 hours (3 days) daytime temperatures will be warm at 22–25°C. This is due to a high pressure system that is expected to disperse clouds and allow the sun to directly warm the surface of the Earth. By contrast, after the sun sets, cool air will move down from higher altitudes and lower the overnight temperature to 12°C or below.

Referencing

There are a number of different referencing styles, such as Harvard or APA. The sources of information must be acknowledged using a referencing style that is suitable for the purpose of the investigation. Check the preferred referencing style or convention of your teacher for in-text referencing and complete a **reference list**. The APA (American Psychological Association) style is the most commonly-used referencing style.

- A bibliography is a list of all the sources used during the research to develop understanding (including information in the journal) even if the information was not used directly or explicitly in the scientific report.
- A reference list only lists the sources used and cited (or in-text referenced) in the scientific report. As such, a reference list is the minimum requirement, or you may choose to extend this to a bibliography. The sources must be listed at the end of the report in alphabetical order (by author last name or organisation name).

Compile your references in a separate document as you conduct the student experiment. This will save you time later. You could note these in your Journal as you work, or copy each URL (website address) as a hyperlink into a digital file which will enable you to easily refer back later, especially if you add your own notes about each. As you browse online it is very easy to lose track of the most useful websites.

Microsoft Word, and other online tools make writing a reference list easier by automatically formatting references in the referencing style of your choice. Always check that this tool has formatted correctly. Many scientific sources now provide an option to cite them, also may give options for the referencing style, when you should choose APA.

TABLE 1.8.6 Examples of citations and references for three common information sources using APA style

Source, information required for reference and example of citation in text	Example of the reference written out in the reference list
article in a scientific journal: • author, initials. (year). Title of article. *Journal title*, *volume number*(issue number), page numbers. • Meagher and Cairns (2014) discovered this genus recently in remote rainforest in tropical Queensland.	Meagher D. & Cairns A. (2014). *Entodontopsis* (Bryophyta: Stereophyllaceae) new to the Australian flora.*Telopea*, *17*, 295–301.
book: • author, initials. (year). *Title of book*(edition, if not first). City: Publisher. • Insects comprise more than 50% of the 1.4 million species described so far (Wilson 1992, page 126).	Wilson E.O. (1992). *The Diversity of Life*. Cambridge: Harvard University Press.
online article: • author, initials. (year). *Title of web page or web document.* Retrieved day, month, year, from URL. • Scientists believe they may have found a cure for white-nose syndrome that is wiping out bats in the United States (Lee 2015).	Lee J.J. (2015). *Killer fungus that's devastating bats may have met its match.* Retrieved 4 September, 2015, from http://news.nationalgeographic. com/2015/05/150527-bats-whitenose-syndrome-treatment-conservationanimals-science.

In-text citations

Each time you write about the findings of other people or organisations, you need to provide an in-text citation and provide full details of the source in a reference list. (Table 1.8.6). In the APA style, in-text citations include the first author's last name and date in brackets (author, date).

The following example shows two types of in-text citation and the reference list entry.

In-text citations
It was reported that in testing of five pro-oxidant additives added to commonly manufactured polymers, none resulted in significant biodegradation after three years (Selke *et al.*, 2015).
Selke *et al.* (2015) reported that in testing of five pro-oxidant additives added to commonly manufactured polymers, none resulted in significant biodegradation after three years.
Reference list entry for the example above would be:
Selke, S., Auras, R., Nguyen, T.A., Aguirre, E.C., Cheruvathur, R., & Liu, Y. (2015), Evaluation of biodegradation—promoting additives for plastics. *Environmental Science & Technology*, 49(6), 3769–3777.

USING AI IN RESEARCH AND WRITING

Generative AI (or just AI) uses chatbots or digital assistants that you can communicate with using fairly natural language to access information in Large Language Models (computer models trained on vast amounts of data). These typically generate text, and some models can generate images, video and audio.

AI can be great for brainstorming ideas about investigations, generating practice questions and generating data for investigations. However, you must follow guidance from your teacher or school on how you can use AI in your work. You could be penalised by your school for using AI-generated material without permission or acknowledgement. It's also important to understand the limitations of AI-generated text. For example, you will be unable to discern the extent of plagiarism or be confident of the original source.

Check with your teacher before using AI in your work.

Acknowledge use of AI using referencing conventions.

Always fact check AI-generated text and references.

Limitations of AI-generated text

Large language models are engineered to match patterns of words, but they can't understand the meaning of your prompt or the output text. They produce text that is similar to patterns of words in the prompts and outputs they have been trained on.

This can result in text that sounds knowledgeable and authoritative but is not fluent or concise when you examine it or try to rephrase it in simple language. AI can 'hallucinate' – that is, make up statements – and this can be obvious or subtle. For example, AI can give correct answers to questions it has generated but give the wrong explanations.

The data that the model is trained on is not necessarily scientifically accurate, so generated text could be incorrect or biased. AI can be prompted to provide references, but these are often not reliable so should always be checked directly at the original source.

AI-generated answers may not model the complete response that aligns with the scope and scale of the syllabus. In class, you will have developed an understanding of the scientific concepts described in the syllabus. In this way you can check the level of information and accuracy provided in an AI-generated response.

Referencing AI sources

You must properly acknowledge the use of AI in your work. Some AI tools provide a URL that will link to generated text. If yours does not, it's a good idea to include the prompt you used and the AI output as an appendix to your work. The following are sample ways to reference AI-generated text (Table 1.8.7).

TABLE 1.8.7 Examples of references for AI-generated text

Referencing style	General format	Example
APA	Author. (Year). Title of software program (Version). Date. URL. Appendix.	OpenAI. (2024). *GPT-4o*. 20 June 2024. URL or 'see Appendix for prompt used and output generated'.
Chicago	Footnote number. AI tool used, Month Day, Year, Creator of tool, URL, Appendix.	1. Text generated using GPT-4o, June 20, 2024, OpenAI, URL or 'see Appendix for prompt used and output generated'.

Prompting the digital assistant

Learning to write effective prompts is essential to get good results from AI. There is no perfect way to write a prompt, but using one of the many frameworks can be useful. Keep the instructions simple—for example, follow these three dot points in your prompt:

- role (Who the chat assistant is being: e.g. policy maker, scientist, consumer)
- task (What output you would like: e.g. an article, summary, set of questions)
- format (What it should look like: e.g. bulleted list, table, graph).

Example: I am a government policy maker working on energy strategy. Give me a one-page summary of pros and cons of using nuclear power in Australia. Consider cost, climate change, security, the environment and First Nations' perspectives in your answer. Present the summary in bullet points and include accurate references for all data.

If necessary, you can refine the results using further prompts. Note that chat assistants are not predictable, and you can get different results using the same prompt.

MULTIMODAL PRESENTATIONS

Student experiments and research investigations are focused investigations resulting in a conclusion related to the initial question posed. In schools, the process occurs over a number of lessons with a time limit but in research science a project can continue for weeks, months or years. The reporting of the final findings is usually a written report published in a scientific journal but can also take place at a scientific conference. In the Queensland Biology syllabus, you are also provided this flexibility in the form of multimodal presentation as an alternative to the written report.

It is important to note that this mode gives you an option for the final presentation of the task only. All criteria and descriptors in the Instrument Specific Marking Guide relating to forming, finding, analysing and interpreting/evaluating need to be met as described in Parts B and C. Multimodal presentations must include at least two modes of delivery at the same time, have a maximum time of 11 minutes and maximum 2000 words. Your two modes are likely to be visual (PowerPoint or poster) and verbal (speech). For average speaking speed, 11 minutes will equate to around 1600–1700 words. Your teacher will need the PowerPoint printed and a printed script - they may record your presentation. Another option is to use the audio record function of PowerPoint. These aspects of assessment will be identified by the school on their endorsed task instructions.

Some considerations regarding the speech are:

- engage and effectively communicate with the audience
- use a clear structure – it is suggested to follow the same outline as a written report (see headings provided in Parts B and C)
- interact with your PowerPoint or poster to point out important aspects
- while it is not advisable to speak on a tangent or set up a conversation with the audience, it would be possible to ask the audience if they have any questions at the end
- use a strong confident tone
- bring your own timer to avoid going over time

Some considerations regarding the visual presentation (for PowerPoint):

- each slide should have a clear focus with minimal text (6–7 lines per slide, 6–7 words per line). Use minimum size 24 font so that audience can read it
- leave time for the audience to read your text rather than reading it word for word. Draw their attention to significant text
- for graphs and tables, present a single one per slide. Leave graph axes or annotations in smaller font and read this information aloud
- use a consistent design, frame or theme for the slides.

Some considerations regarding the visual presentation (for poster):

- use a presentation size large enough to include all your graphs, tables, figures and text
- make the poster 'flow' so the reader clearly follows the investigation from planning through to concluding and evaluating
- allow your audience some time before the presentation to view the information and allow them to formulate questions.

Communicating experimental or research findings is an integral part of a scientist's work. Irrespective of the time span of the investigation, there is a sense of achievement when the results are made known, especially when the process requires you to relate to the topic in depth. By choosing a verbal presentation, you can share this journey with the audience.

1.8 Review

SUMMARY

- Scientific reports use scientific language and representations.
- The language of scientific reports is concise, objective and written in the third person.
- The scientific visual representations include tables, charts, graphs, tables, diagrams, images.
- Visual representations require headings, labels, captions, descriptions, numbering (if more than one item).
- A science journal is a necessary tool for recording detailed information and sources during an investigation.
- Write using your own words to avoid plagiarism.
- Acknowledge sources and any references to the work of others by including a reference list (or full bibliography).
- If using AI for scientific research ensure information is checked against other sources, avoid plagiarism and acknowledge the AI source.
- A written or multimodal format may be used for your report but the same syllabus criteria and descriptors apply to both formats.

KEY QUESTIONS

Describe

1 Clarify the difference between a bibliography and a reference list.

2 Describe what plagiarism is and recognise why it is unacceptable.

Apply

3 List the information that should be recorded about an online source in your journal.

4 Explain the benefit of keeping a scientific journal.

Analyse

5 Rewrite the following texts in your own words.

a A group of patients was administered adrenaline during a cardiac arrest to determine if adrenaline would improve patient outcomes. There was no statistically significant improvement in patient outcome between patients who received adrenaline and those who did not.

b A team of scientists from the Oregon Health and Science University and Korea's Institute for Basic Science successfully repaired a defective gene in human embryos. The gene that was targeted caused a heart defect, known as hypertrophic cardiomyopathy, which causes sudden death in young adults.

c This study found that water temperature had no significant effect on the amount of bacteria remaining on hands after washing. Rather, the length of time spent scrubbing hands, as well as the vigour of scrubbing, had the most effect on removing bacteria from hands.

PART B STUDENT EXPERIMENT

The example for the student experiment report (lipase experiment) used in the text is very comprehensive but if each of the sections were put together the report would exceed the mandated word limit for the task.

The student experiment requires you to undertake the full scientific method. The Queensland Biology General Senior Syllabus 2025 states that this process begins with a practical conducted during class or viewed online. This practical will be modified to conduct your own experiment. It is recommended that during the class practical you record your observations, queries and thoughts as well as collecting and processing data. The record would be an ideal start for your science journal (see Module 1.8 Science journal and Figure 1.8.1). These notes can be used to develop a research question for the student experiment.

The task assesses syllabus objectives, science inquiry skills and relates to student experiment objectives and specifications from the Biology General Senior Syllabus 2025. The student experiment constitutes 20% of the total assessment and may be presented in:

- written form (e.g. scientific report), up to 2000 words, or
- multimodal presentations form (at least two modes delivered at the same time), up to 11 minutes. Refer to Module 1.8 Multimodal presentations.

In the modules that follow, you will find a guide to **scientific method** and an outline for producing a scientific report for the student experiment.

INSTRUMENT-SPECIFIC MARKING GUIDE

Student responses are assessed against an instrument-specific marking guide (ISMG). In developing your experiment and planning your response, it is important to always have in mind the assessment objectives, and in particular the characteristics that are described in the performance level descriptors.

- The ISMG has four criteria. Each criterion carries equal marks:
  - forming
  - finding
  - analysing
  - interpreting and evaluating.
- Criteria have performance levels, against which the qualities of the response are assessed. Table 1.9.1 shows the high-performance level characteristics for the four criteria.
- A performance level consists of a performance level descriptor and performance level mark or mark range.
- The performance level descriptor describes the characteristics that are demonstrated by a response at this quality.

The ISMG criteria, and elaborations of each are shown in Table A, for the highest performances.

Note that in Year 11, this internal assessment task will align with Unit 1 Cells and multicellular organisms and not Unit 3 Biodiversity and the interconnectedness of life. A research project based on IA2, but undertaken in Year 11, will require modification of assessment objectives to relate to Unit 1 subject matter. The ISMG marking guide descriptors and Module references remain the same.

TABLE A The ISMG criteria and highest performance descriptors for marking the scientific report for the student experiment

Criteria	Assessment objectives	Instrument specific marking guide (ISMG) descriptors – Criteria are demonstrated by	Marks	Module
Forming	Describe ideas and experimental findings about describing biodiversity, functioning ecosystems, population ecology or changing ecosystems. Apply understanding of describing biodiversity, functioning ecosystems, population ecology or changing ecosystems. Investigate phenomena associated with describing biodiversity, functioning ecosystems, population ecology or changing ecosystems through an experiment.	• a considered rationale for the experiment • justified modifications to the methodology • a specific and relevant research question • a methodology that enables the collection of sufficient and relevant data • appropriate use of genre and referencing conventions	5	1.10 1.8
Finding	Describe ideas and experimental findings about describing biodiversity, functioning ecosystems, population ecology or changing ecosystems. Investigate phenomena associated with describing biodiversity, functioning ecosystems, population ecology or changing ecosystems through an experiment.	• considered management of risks/ethical issues/ environmental issues • collection of sufficient and relevant raw data • fluent and concise use of scientific language and representations	5	1.11 1.8
Analysing	Apply understanding of describing biodiversity, functioning ecosystems, population ecology or changing ecosystems. Analyse experimental data about biodiversity, functioning ecosystems, population ecology or changing ecosystems.	• correct and relevant processing of data thorough identification of relevant trends/ patterns/relationships • thorough and appropriate identification of the uncertainty and limitations of evidence	5	1.12 1.5 1.6 1.7
Interpreting and evaluating	Interpret experimental processes and conclusions about biodiversity, functioning ecosystems, population ecology or changing ecosystems. Evaluate experimental processes and conclusions about biodiversity, functioning ecosystems, population ecology or changing ecosystems.	• justified conclusion/s linked to the research question • justified discussion of the reliability and validity of the experimental process • suggested improvements and extensions to the experiment that are logically derived from the analysis of evidence	5	1.13 1.7
Total			20	

The final report for the student experiment may be organised under the four criteria headings in the instrument-specific marking guide (ISMG). However, this is not mandated, and you may elect to organise your report in a different but logical order using alternate headings.

Note that items in the Appendix are not included in the word count (up to 2000) and are not used for determining levels of achievement #.

The headings and sub-headings in Table B show one possible structure of a student experiment report (* alternate positioning for the Conclusion section):

TABLE B Possible student experiment structure

Forming	rationale
	research question
	modifications
Finding	risk assessment
	raw data
Analysing	sample calculations
	processed data
	analysis
Interpreting and evaluating	conclusion*
	evaluation of experimental process (reliability and validity)
	improvements and extensions
	conclusion*
	reference list/bibliography
	appendix (if used). (See note above #).

1.9 Forming

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand how to identify and modify an experimental investigation
- be able to research relevant background information that informs the research question
- develop the investigation by forming a research question which includes specific variables
- justify modifications to the original experiment.

RATIONALE

Your student experiment uses a completed class practical or simulation as a basis for further experimentation. This means that the rationale will show an idea flow which begins with an original experimental result and uses scientific reasoning to propose a new and related question (Figure 1.9.1). Often the same or similar experimental apparatus is used but independent and/or dependent variable(s) are altered. Conversely, a new experimental design and equipment may be used to investigate the topic.

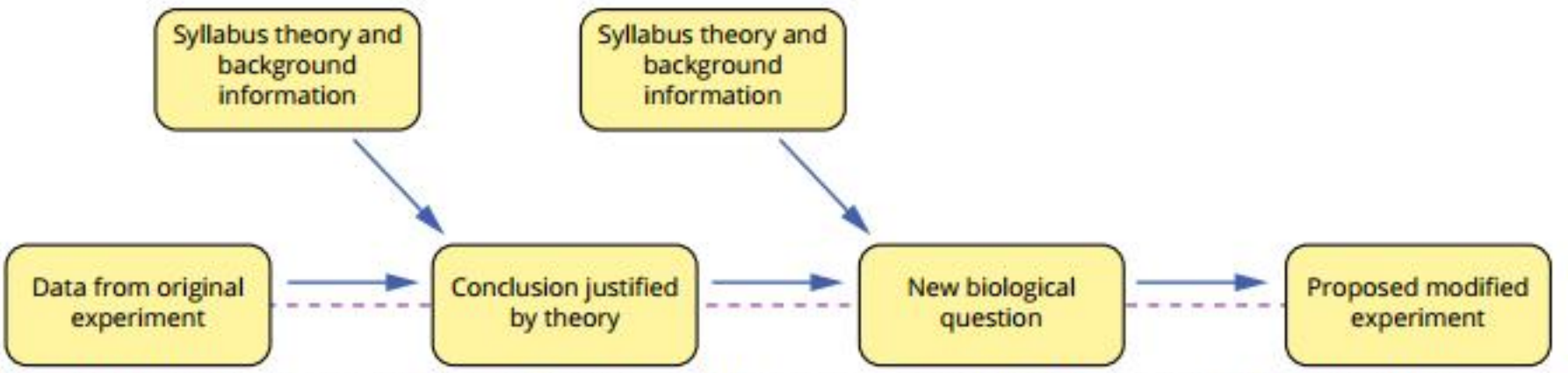

Syllabus theory and background information
Syllabus theory and background information
Data from original experiment
Conclusion justified by theory
New biological question
Proposed modified experiment

FIGURE 1.9.1 Idea flow for reasoning process in developing the Rationale section. A new biological question is developed from biological understanding and the result of the original experiment

Background information

The biological concepts of both the original experiment and the proposed new experiment need to be described. It is important to restrict this to the necessary detail – a full topic explanation or literature review is not required. Make sure that key syllabus concepts are included using relevant biological knowledge and diagrams. Background concepts need to be referenced by scientific literature using in-text referencing. You will need to read widely but are advised to limit your use of biology concepts that are beyond the scope of the syllabus. If your original experiment and proposed new experiment cannot be explained using syllabus concepts from the appropriate unit, you will need to change your experimental focus. While science understanding of Unit 1 and Unit 2 are formative in the concepts of Units 3 and 4, a summative Student Experiment cannot be based on science understanding of Units 1 and 2.

Your explanation of background information demonstrates understanding and knowledge of the direct, and possibly indirect, relationship between the independent and dependent variables and controlled or measured variables.

The syllabus states:

'research relevant background scientific information to inform the modification of the research question and methodology'.

In this syllabus statement we see:

- 'relevant' which means background scientific information relevant to the syllabus as well as relevant to your experiment
- 'inform' which means you have used science concepts to identify a new experiment
- 'modification' the new experiment is related to the first one demonstrated in class, but the variable(s) and/or method are modified
- 'research question' refers to a specific research question with proposed dependent and independent variables
- 'methodology' the details of how you plan to conduct your experiment using scientific method.

Group planning

Parts of your rationale can be conducted with your group – it is appropriate to discuss the findings of the original experiment to be clear about the relationship shown in the data. Also, the group should discuss changes to the method or dependent and independent variables which may be appropriate.

Discuss with your peers the ideas already learnt in class about the topic and possible or expected relationships that could be investigated (Figure 1.9.2). This relationship will be the focus of your research question. In addition, you may propose a refinement of equipment or methodology which will need to be discussed and clarified with your teacher and also reviewed for safety. Check that your school laboratory has the resources for you to perform the intended experiment or investigate the topic. Choose a topic that can provide clear measurable data that shows how variable y depends on variable x.

Consider the following when developing the modified experiment:

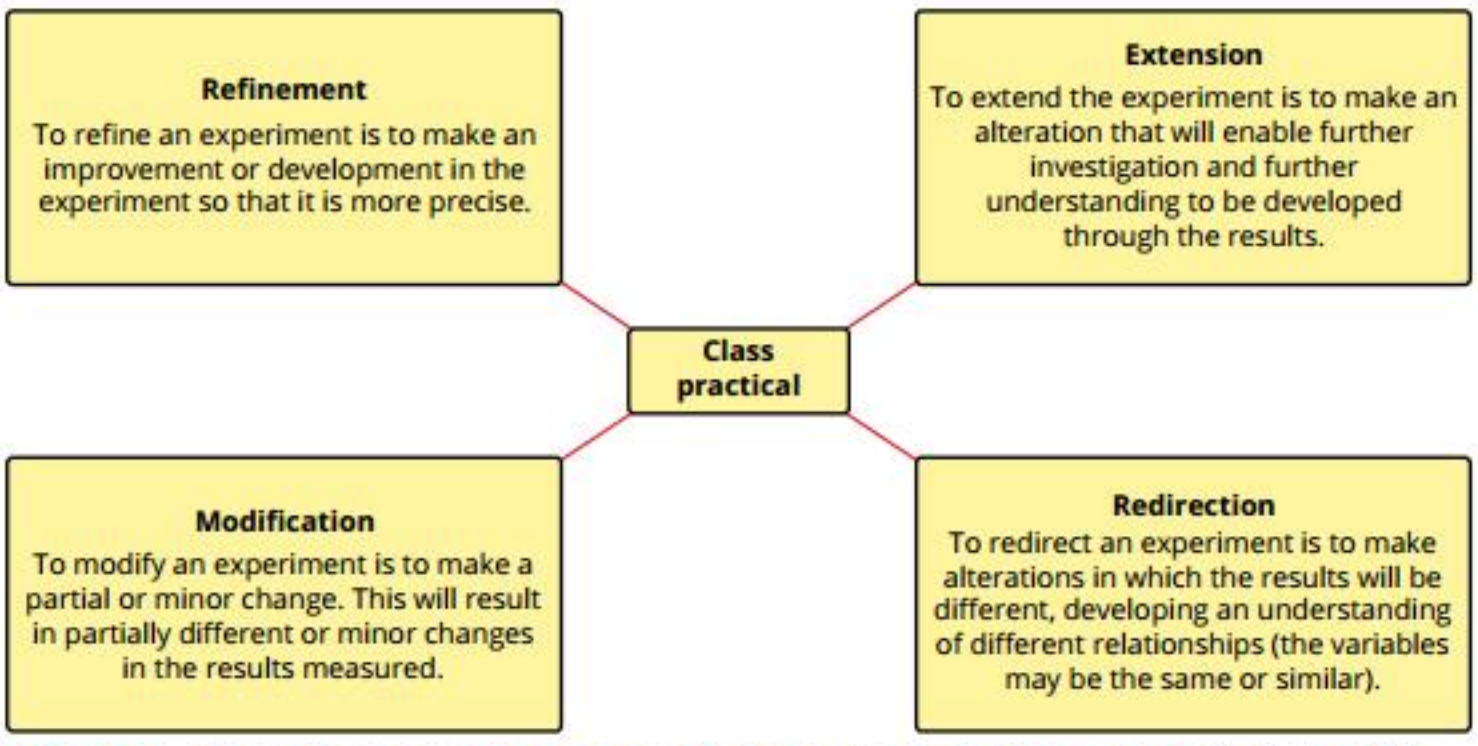

Refinement
To refine an experiment is to make an improvement or development in the experiment so that it is more precise.
Extension
To extend the experiment is to make an alteration that will enable further investigation and further understanding to be developed through the results.
Class practical
Modification
To modify an experiment is to make a partial or minor change. This will result in partially different or minor changes in the results measured.
Redirection
To redirect an experiment is to make alterations in which the results will be different, developing an understanding of different relationships (the variables may be the same or similar).

FIGURE 1.9.2 Changes that can be made to a class practical to develop the student experiment. See Table 1.10.1 'Modifications' below for further information

Table 1.9.1 shows examples of modifying an original experiment into a topic of interest for a student experiment.

TABLE 1.9.1 Examples of modifying a practical into a student experiment

Original experiment	Possible modified experiments
Compare the heights of potted plants grown in soil with fertiliser added, with the heights of plants grown with no fertiliser in their soil.	Is there a difference in potted plant heights grown in the same soil with and without water?
	Is there a difference in potted plant heights grown in the same soil with and without direct sunlight?
	Can the experiment be altered to compare different plant species?
	Can the experiment be altered to compare plant growth at different air temperatures or soil temperatures?
	Can the experiment be conducted to determine the optimum amount of fertiliser for plant growth?

Defining the variables and data collection

The factors that can change during your experiment or investigation are called variables. An experiment or investigation determines the relationship between variables by measuring the results. There are four categories of variables (Figure 1.9.3). You should have only one independent variable. Otherwise, you could not be sure which independent variable was responsible for changes in the dependent variable (the results). Use clearly defined dependent and independent variables, include values for concentrations, and be specific about the number of trials.

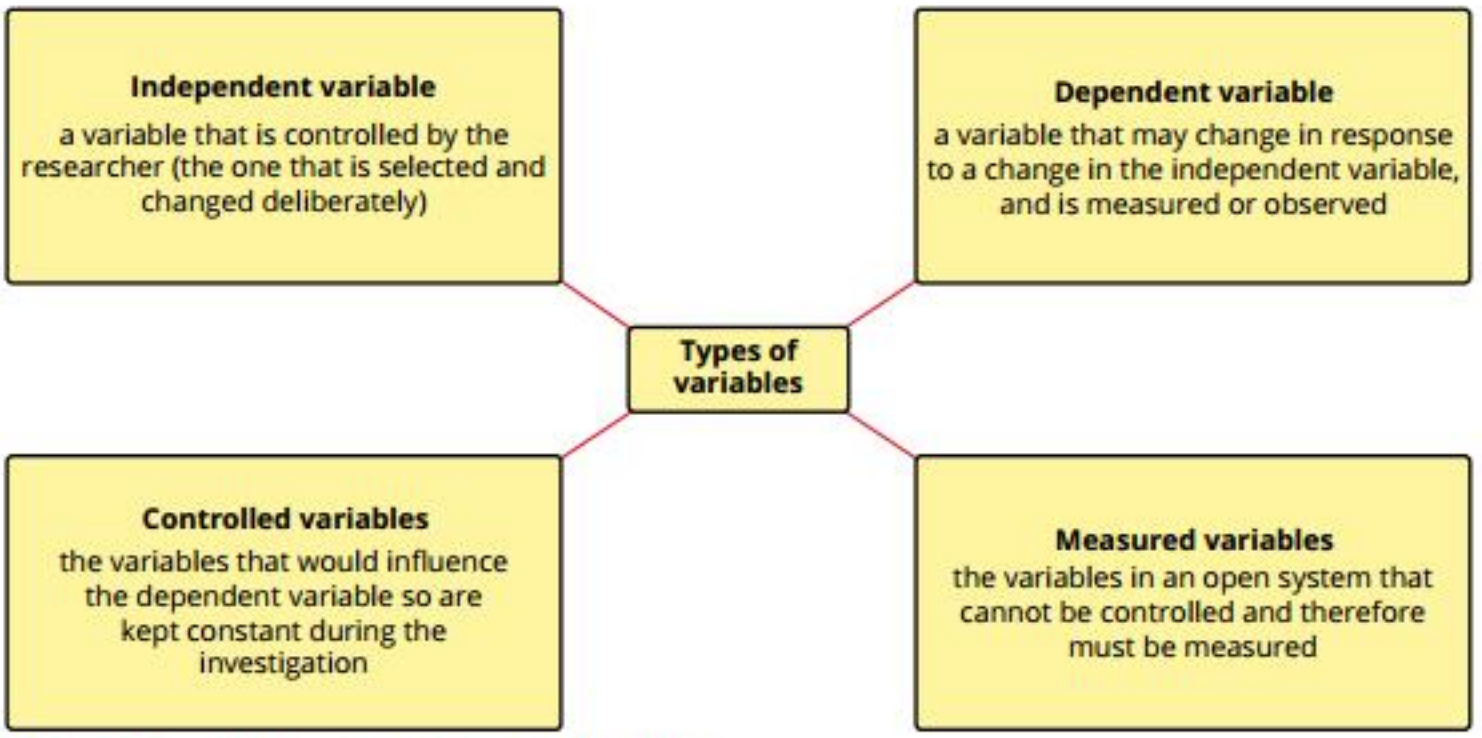

Independent variable
a variable that is controlled by the researcher (the one that is selected and changed deliberately)
Dependent variable
a variable that may change in response to a change in the independent variable, and is measured or observed
Types of variables
Controlled variables
the variables that would influence the dependent variable so are kept constant during the investigation
Measured variables
the variables in an open system that cannot be controlled and therefore must be measured

FIGURE 1.9.3 The four types of variables in experiments

To ensure an investigation is **valid**, it should be designed so that only one variable is being changed at a time. The remaining variables must remain constant so that meaningful conclusions can be drawn about the relationship between variables. In junior science we learn 'change one thing, measure one thing, keep everything else the same'. In your rationale and research question discussions with your group and teacher you will consider:

- the independent variable (the thing that is changed)
- the dependent (measured) variable
- the controlled variables (parts of the equipment, method and conditions that can be kept constant)

- the appropriate raw data that will be collected (quantitative versus qualitative), and how each will be measured, collected or controlled.

Variables and the importance of identifying them correctly, are also noted below in 'Collection of sufficient and relevant raw data'.

Data can either be qualitative or quantitative (Figure 1.9.5).

- **Qualitative data** is descriptive, unmeasurable and uses descriptions or adjectives to record observations. It can, and should be, statistically analysed.
- **Quantitative data** is empirically measurable and uses instruments to record observations as numbers with units.

Qualitative and quantitative data have further subsets in each category (Figure 1.9.4).

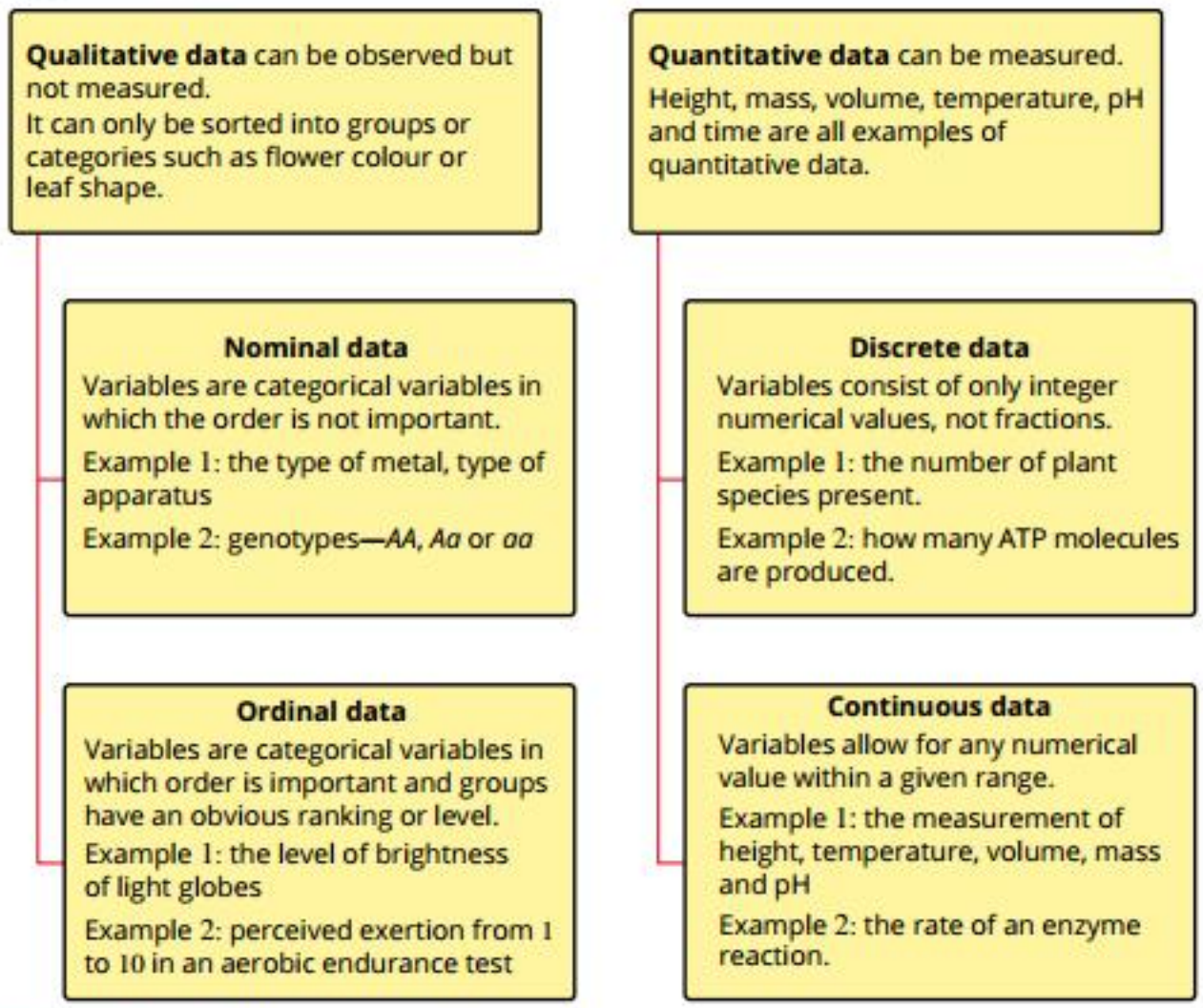

Qualitative data can be observed but not measured.
It can only be sorted into groups or categories such as flower colour or leaf shape.
Quantitative data can be measured.
Height, mass, volume, temperature, pH and time are all examples of quantitative data.
Nominal data
Variables are categorical variables in which the order is not important.
Example 1: the type of metal, type of apparatus
Example 2: genotypes—*AA*, *Aa* or *aa*
Discrete data
Variables consist of only integer numerical values, not fractions.
Example 1: the number of plant species present.
Example 2: how many ATP molecules are produced.
Ordinal data
Variables are categorical variables in which order is important and groups have an obvious ranking or level.
Example 1: the level of brightness of light globes
Example 2: perceived exertion from 1 to 10 in an aerobic endurance test
Continuous data
Variables allow for any numerical value within a given range.
Example 1: the measurement of height, temperature, volume, mass and pH
Example 2: the rate of an enzyme reaction.

FIGURE 1.9.4 Quantitative and qualitative variables

Measurement of temperature requires an instrument and provides quantitative data whereas sensing the production of heat by touch provides qualitative data (Figure 1.9.5).

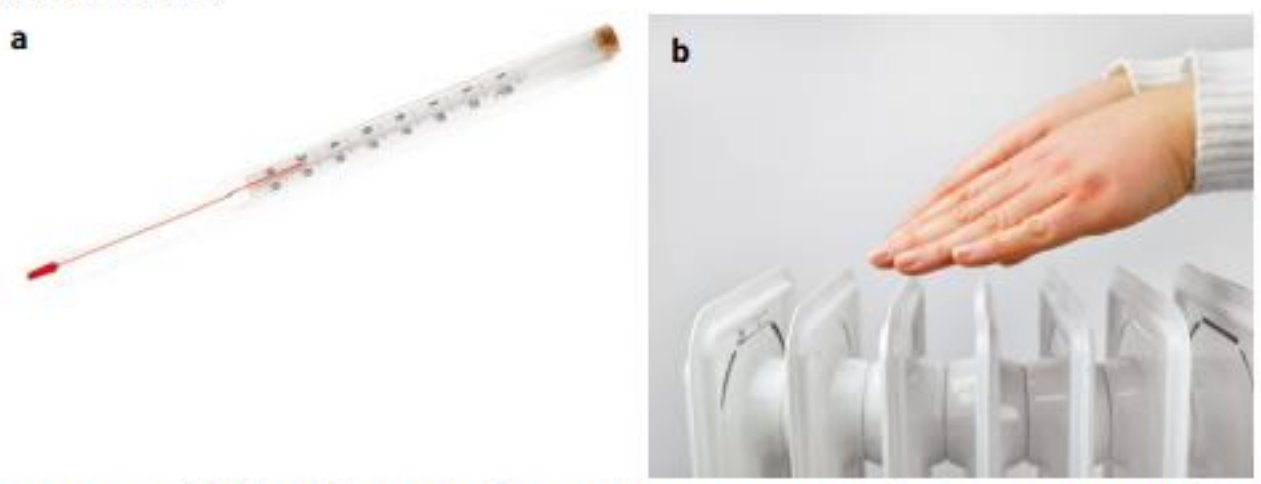


FIGURE 1.9.5 (a) A liquid thermometer for quantitative measurement of water temperature provides numerical values which can be statistically analysed. (b) Feeling heat radiating from a heater is an example of qualitative data as it is based on personal observation (e.g. cold, warm, hot). Qualitative data cannot be statistically analysed.

Depending on the experiment, it may be appropriate to record the qualitative observation (e.g. colour change or precipitate formation during a chemical food test), or to measure the quantitative results (e.g. volume of oxygen gas produced or a pH measurement), or sometimes to do both.

Sourcing appropriate equipment and materials

You will need to decide on the materials, technology and instrumentation that will be used to carry out your experiment or investigation. It is important to find the right balance between items that are easily accessible and those that will obtain accurate and precise results. The precision of the chosen instrumentation affects the level of measurement uncertainty and is recorded as a ± value in the raw data table (refer to Module 1.5). Instrumentation choices are described in 'Justified Modifications to the Methodology'.

Control group

It is difficult to control every aspect and condition in an experiment; to overcome this, a second group is often used within the experiment. This is called a **control group** that is identical in every way to the first group (the experimental group) except for the single experimental (independent) variable that is being tested. In practice the experimental group will have a range of independent variables applied, but the control group has no treatment at all. It could be described as the 'blank' or 'zero' group. This allows the further examination of effect of the independent variable on the dependent variable and identifies the possible effect of outside factors (uncontrolled variables) on the data collected.

For example, an experiment is conducted to test the effect of an antimicrobial substance on bacterial lawn across an agar plate. The experimental group would be agar plates with an even spread of bacteria (*E. coli*) and the antimicrobial agent added. One control group would test that unopened agar plates are sterile from the start without any bacteria or antimicrobial substance being introduced. A second control group would test sterile water on the bacterial lawn plate instead of the antimicrobial substance (Figure 1.9.6).

FIGURE 1.9.6 Gloves are worn to handle a Petri dish covered with a microbiological culture of *E.coli* bacteria.

Another example is an investigation in which different concentrations of enzyme solutions are added to samples of hard-boiled egg white. The control group would use water instead of an enzyme solution.

Sample size

Because a single measurement or experimental result could be affected by errors, replication of samples within an experiment and repeat trials are key components of reliability. A larger sample size will improve:

- the representation of the phenomenon
- identification of natural variation, errors and uncertainty
- the results by offering more evidence to support the experimental results
- repeatability and therefore reliability.

Considering the limited time available for planning, conducting, and reporting the experiment, a practical number of trials is essential. Ten hours are dedicated to writing the reports. To ensure data reliability, at least three trials per independent variable are recommended. A reasonable number of trials might be 15, consisting of either five data points with three measurements each or three data points with five measurements each. The number of independent variables can vary based on the expected relationship: three variables might suffice for linear relationships, while five data points are generally recommended for logarithmic or non-linear relationships. This issue is further described in 'Collection of sufficient and relevant raw data' (page e105– e108).

WRITING A RESEARCH QUESTION

When you have decided on a dependent variable, independent variable, controlled variable(s) and method of testing, you are ready to write your research question.

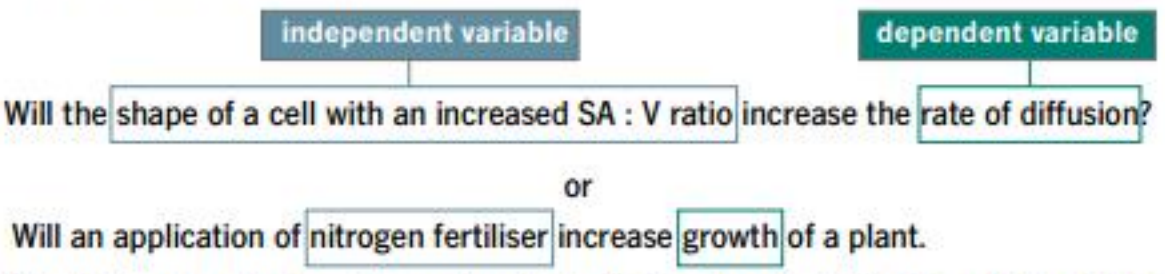
A research question directs and describes the scientific inquiry activity. Its purpose is to focus the student experiment, inform the direction of the research, and guide all stages of inquiry, analysis, interpretation and evaluation.

The question determines the experiment, and the experiment tests the question. A research question should:

- be specific and relevant to the class experiment
- clearly identify the subject matter of the experiment
- specify the scope or conditions of the inquiry
- aim to find trends or patterns of relationships between two variables.

A research question should state the independent and the dependent variables. It should ask if the independent variable will affect the dependent variable. The research question must specifically outline variables in ways that are measurable; this also allows the research question to be answered.

Look at the two examples below:


independent variable
dependent variable
Will the shape of a cell with an increased SA : V ratio increase the rate of diffusion?
or
Will an application of nitrogen fertiliser increase growth of a plant.

The first example is well-constructed – it has clearly defined variables that can be measured.

The second example is not as well-constructed. It does not have enough detail about the nitrogen fertiliser (there are several kinds) and does not say how plant growth will be measured – for example, height of the main stem or the number of new leaves. The second research question has potential but needs refinement (Figure 1.9.6).

A well-constructed research question should have the following characteristics.

- It should include measurable variables—the independent variable and the dependent variable.
- It should have a guiding word, such as *how*, *what* or *will*.
- It should be phrased so that a definitive answer can be developed.
- It should state key (not all) controlled variables or conditions.
- It can state a proposed relationship.

Table 1.9.2 gives examples of constructed research questions.

TABLE 1.9.2 Structuring a research question

Guiding word(s)	Example research questions
will	*Will* natural antibiotics be more effective than synthetic antibiotics?
how	*How* does the volume of cells of the same shape affect diffusion? *How* does substrate concentration affect enzyme activity? *How* do different wavelengths of light influence the number of chloroplasts in plant cells?
can	*Can* enzyme concentration increase the rate of enzyme activity?
do/does	*Does* surface area or volume affect the rate of diffusion?
should	*Should* hands be washed with plain soap or antibacterial soap?
what	*What* difference does surface area make to diffusion?
why	*Why* do cells in different layers of a plant leaf have different concentrations of chloroplasts? *Why* is the relationship between enzyme activity and temperature non-linear?

Evaluating your research question

Once a research question has been developed, stop to evaluate it before progressing. Follow the prompts in Figure 1.9.7 to refine and improve it.

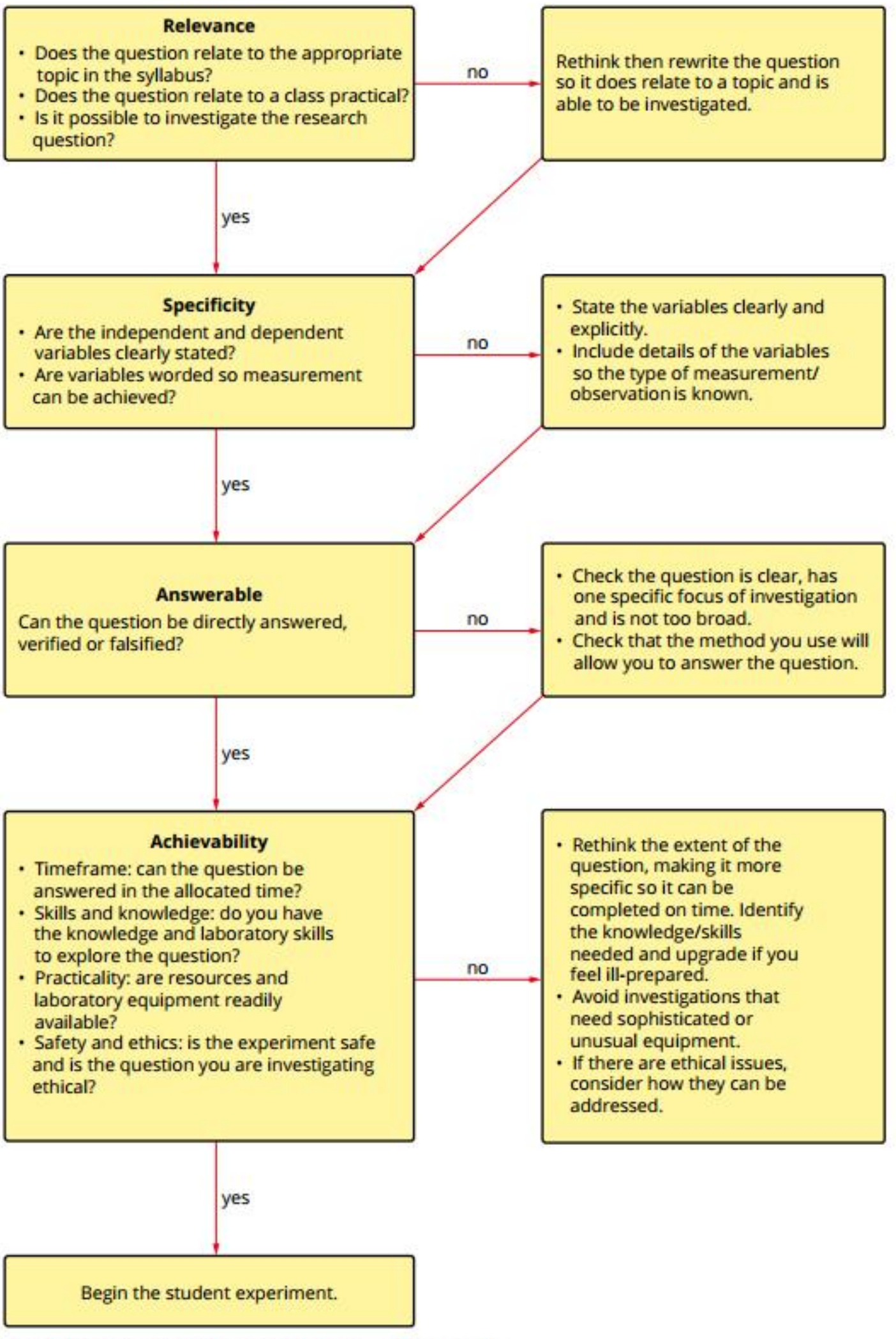

Relevance
• Does the question relate to the appropriate topic in the syllabus?
• Does the question relate to a class practical?
• Is it possible to investigate the research question?
no
Rethink then rewrite the question so it does relate to a topic and is able to be investigated.
yes
Specificity
• Are the independent and dependent variables clearly stated?
• Are variables worded so measurement can be achieved?
no
• State the variables clearly and explicitly.
• Include details of the variables so the type of measurement/ observation is known.
yes
Answerable
Can the question be directly answered, verified or falsified?
no
• Check the question is clear, has one specific focus of investigation and is not too broad.
• Check that the method you use will allow you to answer the question.
yes
Achievability
• Timeframe: can the question be answered in the allocated time?
• Skills and knowledge: do you have the knowledge and laboratory skills to explore the question?
• Practicality: are resources and laboratory equipment readily available?
• Safety and ethics: is the experiment safe and is the question you are investigating ethical?
no
• Rethink the extent of the question, making it more specific so it can be completed on time. Identify the knowledge/skills needed and upgrade if you feel ill-prepared.
• Avoid investigations that need sophisticated or unusual equipment.
• If there are ethical issues, consider how they can be addressed.
yes
Begin the student experiment.

FIGURE 1.9.7 Steps to refine and improve the research question

Formulating a hypothesis

A hypothesis may be developed. A hypothesis is a statement that proposes a relationship between variables and an expected result of the experiment – it is based on some level of understanding. Having completed extensive background research and with knowledge of the syllabus (rationale), you will have the means and understanding to state the expected results in the form of a hypothesis.

The student experiment does not necessarily require a hypothesis, and it may not always be appropriate or beneficial. Because the hypothesis proposes a specific relationship between the independent and dependent variables, the hypothesis can either be supported or refuted by the results. Note that a hypothesis can be supported or confirmed, but not 'proved', whereas it can be disproved (refuted). Figure 1.9.8 shows key components of a hypothesis.

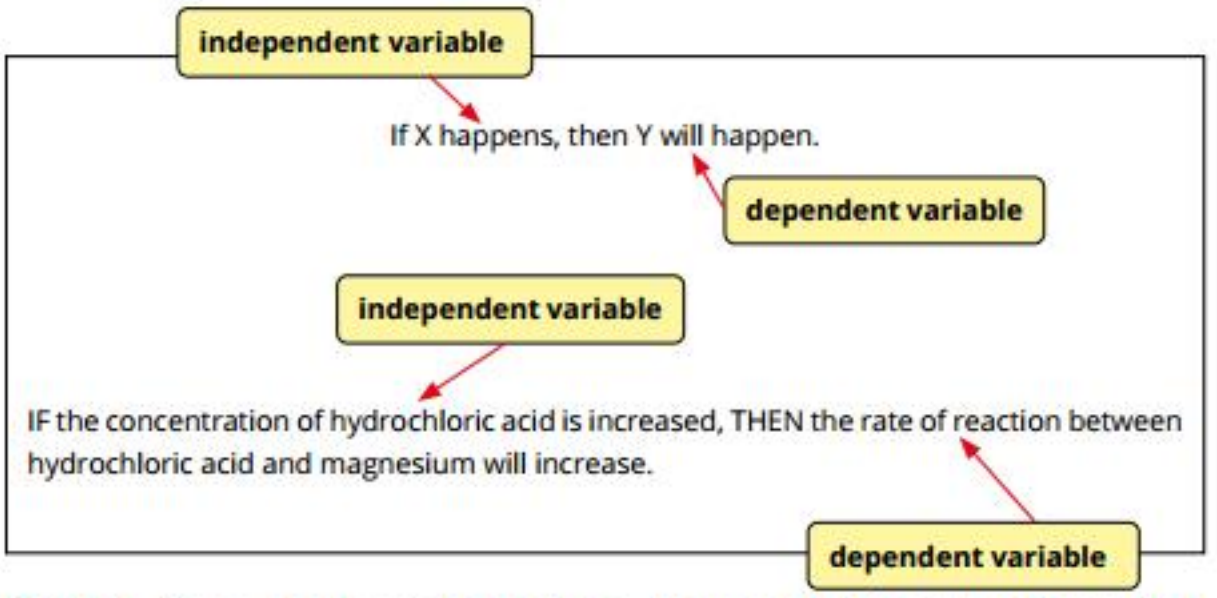

independent variable
If X happens, then Y will happen.
dependent variable
independent variable
IF the concentration of hydrochloric acid is increased, THEN the rate of reaction between hydrochloric acid and magnesium will increase.
dependent variable

FIGURE 1.9.8 The key components of a hypothesis— in terms of the dependent and independent variables

The following examples (Table 1.9.3) demonstrate the relationship between a hypothesis and a research question.

TABLE 1.9.3 Research question examples and associated example hypotheses

Research question	Hypothesis
Is there a linear relationship between exercise intensity and heart rate?	As the intensity of exercise increases, the heart rate will increase in a linear relationship.
Will plant leaves wilt when room temperature increases from 25°C to 55°C?	The degree of wilting in plant leaves will increase as room temperature increases from 25°C to 55°C.
Does 20% tea tree oil have antibacterial properties?	A tea tree oil solution of 20% will result in a larger zone of inhibition on agar plates with *E.coli* lawn culture, than on the control with water.

A research question is a required component of the Student Experiment.

A hypothesis defines the expected link between dependent and independent variables and supports the research question but is not an assessed component of the ISMG. It is a useful but optional inclusion, since it may not always be possible to write a hypothesis for every research question.

JUSTIFIED MODIFICATIONS TO THE METHODOLOGY

The previous section shows how complex the forming (planning) part of the Student Experiment is. You, with your group and teacher, will have decided on key aspects of the modified experiment – independent, dependent and controlled variables, identified a procedure and equipment for use and written a research question. It is necessary to document each modification to the original experiment in your report and justify each proposed modification.

The methodology of your experiment is a specific step-by-step procedure that your teacher will require you to prepare prior to the experiment. However, full detail is generally not required in the final report. In the modifications section, write an overview of the original method (use an in-text reference) in paragraph form and provide a written or tabulated description of the modifications proposed. Include specific details about instruments, chemicals (with concentrations), techniques employed and number of trials. Each modification is justified by stating the type of change ('refine', redirect' or 'extend') and how it will improve the investigation. If possible, link each modification to random and systematic error. Modification examples related to the research questions in Table 1.9.3 are shown in Table 1.9.4.

TABLE 1.9.4 Example modifications to an original experiment with justification

	Modification	Justification
Refinement	Using a heart rate monitor instead of a wrist pulse count.	• Allows for more precise readings, resulting in a more accurate relationship and decreases measurement uncertainty. • Reduces possible systematic error.
Refinement	Collection of more data – five trials each with 10 plants rather than one.	• Enables the identification of uncertainty of evidence and trends produced from data.
Redirection	Observe wilted leaves under a binocular microscope against a grid, before and after temperature exposure.	• Investigates the changes in a leaf that has wilted more closely.
Extension	Measuring a range of different plants for leaf wilting. Investigate a range of concentrations of tea tree oil on bacterial lawn plates.	• Understanding how different plants respond to increased temperature. • To find the most effective concentration of tea tree oil to use as an antiseptic.

One way to modify an original experiment is to improve the precision of the laboratory equipment. With higher-precision instruments, there is less chance of error and lower uncertainty in the measurement. Table 1.9.5 demonstrates the benefits of greater-precision instruments through lower measurement uncertainty.

TABLE 1.9.5 The difference in measurement uncertainty between instruments: (a) a glass thermometer with a precision of ±2.5°C; (b) a measuring beaker with a precision of ±5 mL; (c) a digital laser thermometer with a precision of ±0.2°C; and (d) a measuring cylinder with a precision of ±0.25 mL

Lower-precision instruments	Higher-precision instruments
A	C
B	D

Putting it all together

The sample experiment report below follows the ISMG criteria for the Forming section. It covers rationale, research question and justified methodology for the experiment. This may be used as a reference tool for writing your own report. Note that this sample uses syllabus understanding from Unit 1 Cells and multicellular organisms. Students planning a Year 12 experiment report must use syllabus concepts from Unit 3.

Effect of lipid and triglyceride concentration in different milk types on lingual lipase efficiency.

Rationale

The original experiment focused on the transformation of a carbohydrate (lactose) in milk into simple sugars using the enzyme, amylase, and how temperature (independent variable) impacted the rate of reaction. The experiment was found to have limitations: there was minimal lactose breakdown by amylase, which resulted in a barely measurable change in pH (dependent variable). To modify this, the experiment will be redirected to focus on the reaction catalysed by lingual lipase enzyme with the fats in milk as substrate. Instead of focusing on temperature and rates of reaction, this will be shifted to extending the types of milks being tested to include Full Cream, Almond, Soy, Lite, and Skim milk (independent variable). The temperature will be refined to 37°C and a time limit will be set to 10 minutes per trial. The trials will still be measured by the change in pH (dependent variable).

Lingual lipase is a digestive enzyme that is secreted at the base of the tongue. Digestion of lipids and fats begins in the mouth by the action of the lingual lipase. Contrary to other digestive lipases (e.g. pancreatic lipase), lingual lipase can effectively penetrate the milk fat globule to initiate the digestive processes, successfully breaking the ester bonds to form free fatty acids (FFA) and partial glycerides in a hydrolysis reaction (Stephen, 2020). The reaction is a hydrolysis one and catabolic because it uses water as a reactant and breaks down lipids into smaller molecules. In humans, lingual lipase is present in von Ebner glands of the tongue and functions optimally at homeostatic conditions. Research has indicated that lingual lipase denatures when conditions do not reach, or exceed, a temperature of 37°C and a pH between 3.0–6.0, as the active site of the enzyme begins to change (Adair, 2022).

Lipids are fatty, waxy, or oily compounds that are essential structural components of all cell membranes. They contribute to some of the body's vital processes and the homeostatic function (Ahmed, Shah & Ahmed, 2022). Lipolytic activity was assayed using five different milks as a substrate to ensure a wide range of data can be collected.

The types of milk that were investigated (full cream, skim, lite, soy, and almond) were selected based upon the amount of fat present in each milk solution. To measure the efficiency of lingual lipase, a 5 mL sample was added to 240 mL of each milk, where the pH of the solution could be recorded. As the hydrolysis reaction occurs, the carboxylic acid group (of the FFA) can donate hydrogen ions, increasing the hydrogen ion concentration in the milk solution (Stephen, 2020). The increase in hydrogen ion concentration lowers the pH of the solution, therefore making it a valid measure of the efficiency of the lingual lipase. In theory, research indicated that a greater decrease in pH suggests a more efficient catalysation. It was determined that a greater rate of reaction would be a favourable outcome as it can be used to further develop future studies and trials.

Overall, the aim of the modified experiment was to test how changing the substrate would impact the rate of reaction (efficiency) catalysed by lingual lipase. The targeted substrate found in milk is lipids and triglycerides, of varying size and chemical composition. As such, any five milk types could be investigated if a contrast in fat content was evident.

Research question

How does changing the lipid concentration of a milk (full cream, lite, soy, almond and skim) impact the efficiency (rate of reaction) of lingual lipase when tested at 37°C, over a 10-minute period?

Experimental

The original experiment used amylase as a catalyst in a reaction with starch to observe the effect of temperature on the rate of reaction. The experiment focused on the transformation of carbohydrates into sugars, and as such was altered. The modifications and justification for each method are outlined in Table 1.

TABLE 1 Forming the new methodology

Original method	Modification	Type of modification	Justification
Amylase was used as a catalyst.	using lipase instead of amylase	redirect	to improve the reliability and validity of the data being collected as pH change in fats was deemed a valid measure of enzyme performance
Only one milk was tested.	using a sample size of five types of milk (full cream, lite, soy, almond and skim)	extension	modified from a sample size of one to improve accuracy and observe trends more effectively
Temperature was recorded as hot, warm and cold.	using a water temperature of 37°C measured accurately	refinement	to ensure a controlled temperature throughout all trials to keep a fair test, and improve accuracy
One sample was measured per temperature.	three trials for each of the five milk samples	extension	to ensure sufficient data is collected and that the experiment remains fair.
No time limit was set.	introducing a 10-minute limit per trial	refinement	refined from no limit to ensure that the milks can be compared, trends calculated, and the experiment is conducted accurately

1.10 Finding

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- conduct a risk assessment
- undertake an experiment encompassing sufficient and relevant data
- collect and present data.

RISK ASSESSMENT

When planning an experiment, it is important for your safety and the safety of others that you consider the potential risks. Risks are potential negative outcomes from carrying out an activity. Everything we do has some risk involved. **Risk assessments** are performed to identify, assess and control risks. A risk assessment should be performed for any situation, whether in the laboratory or out in the field, that could cause harm to people or animals.

The first step in the risk assessment process is to assess all risks in the environment in which the experiment or fieldwork will be carried out. This is the inherent level of risk. The second step is to identify control measures that will reduce the inherent level of risk to low risk. For example, glass equipment in the laboratory is a hazard as it can cause injury if it breaks. Injury is a negative outcome and so it is a risk. A control measure to reduce this risk is handling the glass equipment carefully.

Your report should outline risks, include details such as the names and concentrations of any chemicals, and how the risks will be managed. The key consideration is for the personal safety of the people in the laboratory. However, you also need to consider and include a statement about the ethics of the investigation and how potential effects on the environment are considered and managed.

Your risk assessment will have three sections (Table 1.10.2):

- hazard—the name of the item or activity that could cause risk
- risk—the possible negative outcome that could occur
- control measures—the practices or processes to avoid the risk.

TABLE 1.10.1 GHS pictograms used as warning symbols on chemical labels

GHS pictogram	Use	GHS pictogram	Use	GHS pictogram	Use
	flammable liquids, solids and gases; including self-heating and self-igniting substances		oxidising liquids, solids and gases, may cause or intensify fire		explosion, blast or projection hazard
	corrosive chemicals; may cause severe skin and eye damage and may be corrosive to metals		gases under pressure		fatal or toxic if swallowed, inhaled or in contact with skin
	low level toxicity; this includes respiratory, skin and eye irritation, skin sensitisers and chemicals harmful if swallowed, inhaled or in contact with skin		hazardous to aquatic life and the environment		chronic health hazards; this includes aspiratory and respiratory hazards, carcinogenicity, mutagenicity and reproductive toxicity

Often, the original experiment will have used similar equipment and so information from its risk assessment can be used. Check with your teacher or laboratory technician regarding new equipment that you are planning to use.

To identify risks, think about the following:

- Laboratory rules of the school. For example, procedures for behaviour in a laboratory and procedures for equipment use.
- The activity that you will be carrying out, including physical issues such as lifting heavy objects, handling sharp or hot or contaminated objects, and how you will use equipment like glassware, digital devices and electrical equipment, or any chemicals that you will be handling. Consider your sources of heat and electricity.
- The facilities of the laboratory. These include emergency equipment on site, ventilation of the laboratory, facilities such as a fume hood and if the physical space available is suitable and safe for your work.
- The chemical codes and SDS data (Safety Data Sheet) for chemicals in use. Consider also concentrations of chemicals, hazards associated with products of chemical reactions and safe disposal of chemicals.
- What protective clothing (PPE) you should wear e.g. lab coat, disposable gloves and goggles.

Chemical codes

The chemicals at school or in a hardware shop have warning symbols on the label. These are a chemical code indicating the nature of the contents. From 2017, the Globally Harmonised System of Classification and Labelling of Chemicals (GHS) was introduced in Australia for use in workplaces, including school laboratories. GHS revisions are published from time to time. Your teacher and laboratory technician can advise of any relevant updates. Some common pictogram codes and their meanings are shown in Table 1.10.1.

Safety Data Sheets

Each chemical substance has an accompanying document called a **Safety Data Sheet (SDS)** (Figure 1.10.1). An SDS contains important safety and first aid information about each chemical you commonly use in the laboratory. The SDS provides employers, workers and health and safety representatives with the necessary information to safely manage the risk of hazardous substance exposure.

All schools store SDS information for materials in use in laboratories and these must be consulted when planning investigations. Online platforms such as RiskAssess allow quick access to these databases for a planned experiment. In these types of software programs, the potential hazards for each aspect of the experiment are provided when proposed equipment and materials are listed. Handling procedures are also suggested.

1. IDENTIFICATION OF THE MATERIAL AND SUPPLIER

Product Name: **HYDROCHLORIC ACID - 20% OR GREATER**

Recommended use of the chemical and restrictions on use: Precursor for generation of chlorine dioxide gas used in water treatment.

Supplier: Ixom Operations Pty Ltd
ABN: 51 600 546 512
Street Address: Level 8, 1 Nicholson Street
Melbourne 3000
Australia

Telephone Number: +61 3 9665 7111
Facsimile: +61 3 9665 7937
Emergency Telephone: **1 800 033 111 (ALL HOURS)**

Please ensure you refer to the limitations of this Safety Data Sheet as set out in the "Other Information" section at the end of this Data Sheet.

2. HAZARDS IDENTIFICATION

Classified as Dangerous Goods by the criteria of the Australian Dangerous Goods Code (ADG Code) for Transport by Road and Rail; DANGEROUS GOODS.

This material is hazardous according to Safe Work Australia; HAZARDOUS SUBSTANCE.

Classification of the substance or mixture:
Corrosive to Metals - Category 1
Skin Corrosion - Sub-category 1B
Eye Damage - Category 1
Specific target organ toxicity (single exposure) - Category 3

SIGNAL WORD: DANGER

Hazard Statement(s):
H290 May be corrosive to metals.
H314 Causes severe skin burns and eye damage.
H335 May cause respiratory irritation.

Precautionary Statement(s):

Prevention:
P234 Keep only in original container.
P260 Do not breathe mist / vapours / spray.
P264 Wash hands thoroughly after handling.
P271 Use only outdoors or in a well-ventilated area.
P280 Wear protective gloves / protective clothing / eye protection / face protection.

FIGURE 1.10.1 Extracts of a Safety Data Sheet (SDS) for concentrated hydrochloric acid. The SDS alerts the reader to any potential hazards when using a substance, including appropriate measures to reduce risk of harm

It is important to note that to address the syllabus requirement to show 'considered management of risk', you need to do more than download an SDS or use the output of risk assessment software. For safety, you must practise the control measures to reduce risk as you conduct your experiment. The syllabus specification requires you to 'conduct a risk assessment and account for risks in the methodology.'

This means that, as described above, the following three factors need to be aligned and clearly presented in the report:

- hazard
- risk (possible negative outcome)
- control measures.

This can be done by writing a paragraph or preparing a table of your own (Table 1.10.2). The supporting SDS or files from risk assessment software, should be placed in the Appendix and key facts from them used in your report.

TABLE 1.10.2 Sample risk assessment table showing hazards, risks and control measures to prevent the risks

Risk assessment		
Hazard	**Risk**	**Control measures**
Glass equipment	Breakage of glass	Cautiously handle
		Stay aware of surroundings
		Listen to teachers' directions
1 M copper sulfate	Skin and eye irritation	Gloves, apron, goggles
1 M potassium nitrate	Eye irritation	Goggles
1 M zinc nitrate	Inhalation: light headedness, respiratory irritation	Face mask
		Handle powder gently
Zinc metal	Cuts	Cautiously handle
Copper metal	Skin irritation	Gloves

Risk controls

The hierarchy of risk controls is shown in Figure 1.10.3. It is organised from the most effective to least effective. The most commonly used risk control measure that addresses the majority of risks is personal protective equipment (PPE). The least common but most protective control measure is eliminating the risk from the scientific investigation.

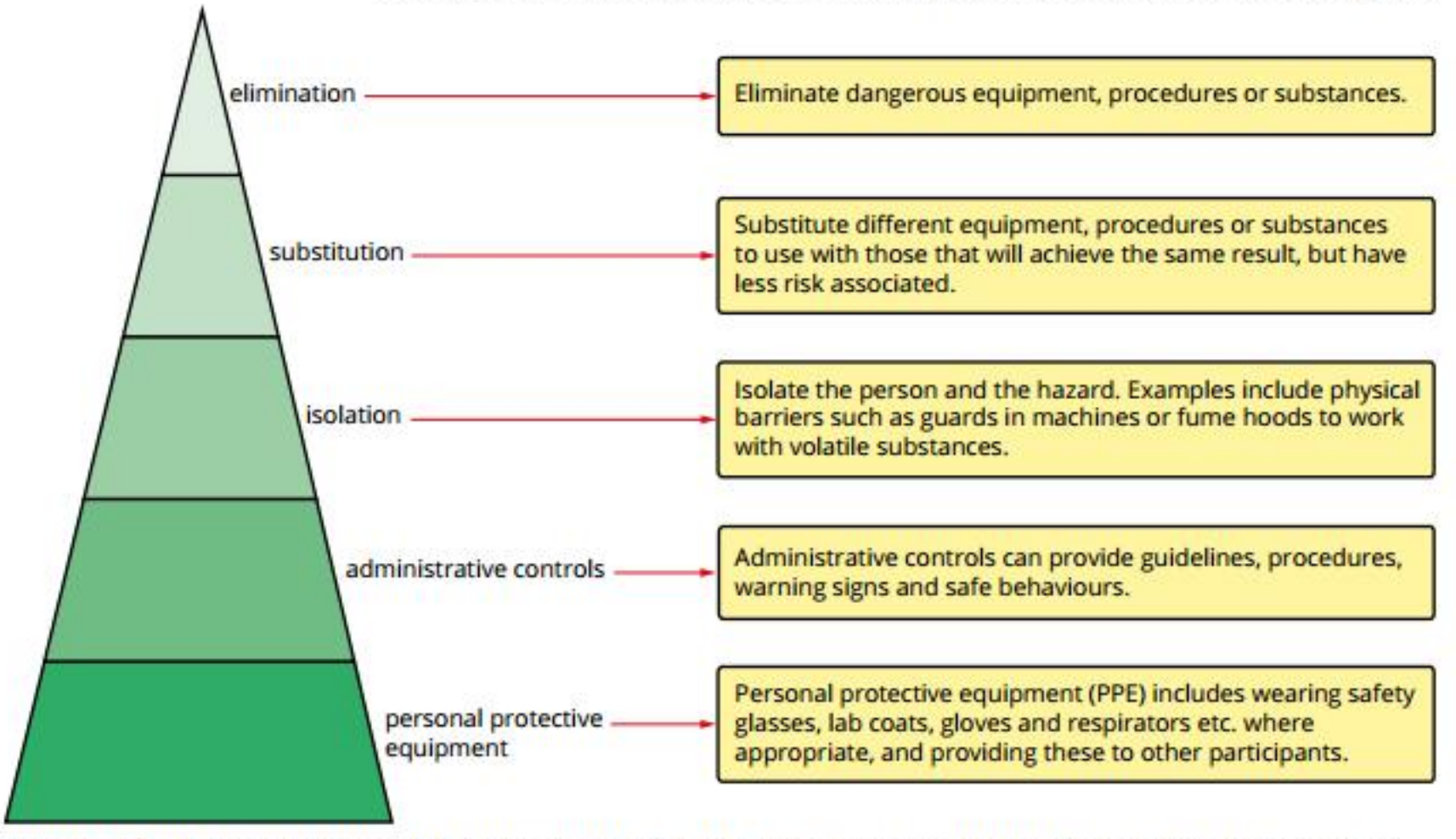

elimination
Eliminate dangerous equipment, procedures or substances.
substitution
Substitute different equipment, procedures or substances to use with those that will achieve the same result, but have less risk associated.
isolation
Isolate the person and the hazard. Examples include physical barriers such as guards in machines or fume hoods to work with volatile substances.
administrative controls
Administrative controls can provide guidelines, procedures, warning signs and safe behaviours.
personal protective equipment
Personal protective equipment (PPE) includes wearing safety glasses, lab coats, gloves and respirators etc. where appropriate, and providing these to other participants.

FIGURE 1.10.3 The hierarchy for risk control is shown in this pyramid, marked from bottom to top in order of increasing importance.

Personal protective equipment

Everyone who works in a laboratory wears items that help keep them safe, such as those shown in Figure 1.10.4. These items are called **personal protective equipment (PPE)** and include:

- safety glasses/goggles
- shoes with covered tops
- disposable gloves when handling certain chemicals
- a disposable apron or a lab coat if there is risk of damage to clothing
- hair net or hair tied back for using a Bunsen burner.

FIGURE 1.10.4 Examples of PPE shown are protective eye wear, lab coats and gloves.

Environmental and ethical considerations

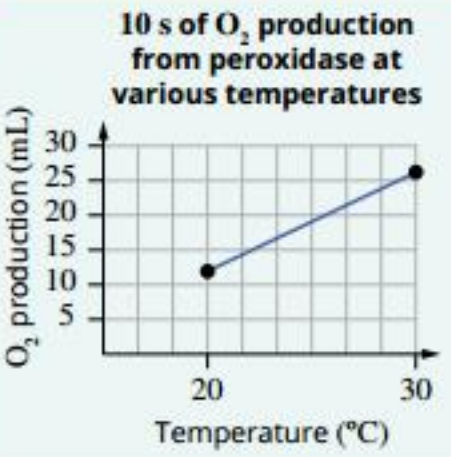
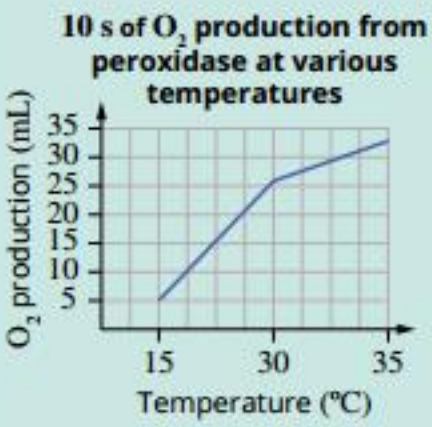
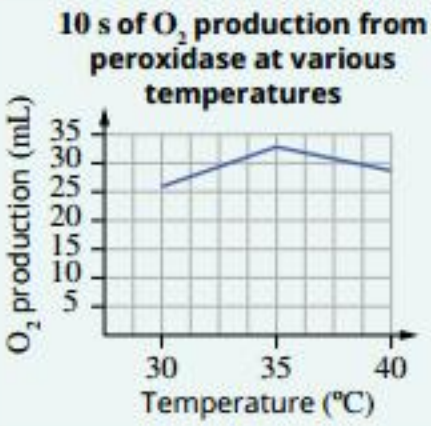
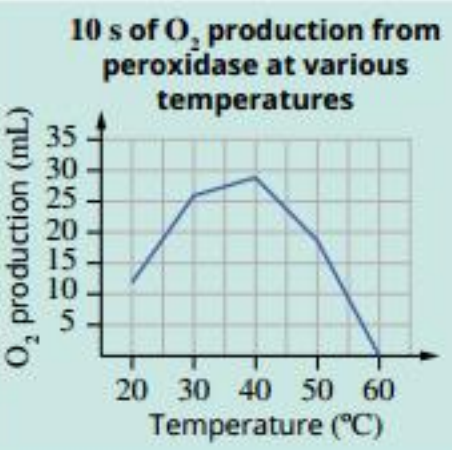
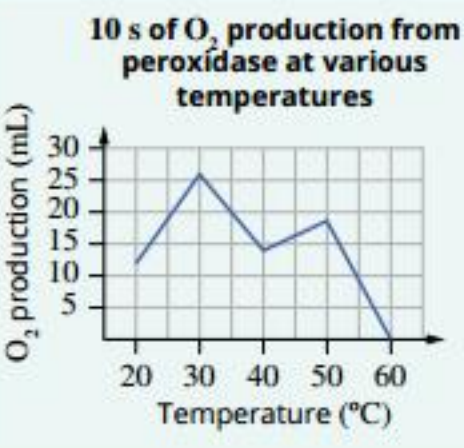
When planning an investigation, identify all possible environmental and ethical considerations, and evaluate mitigation of these issues. Reactants and products of chemical reactions are often toxic to aquatic organisms. Because wastewater will eventually re-enter the natural environment, many common laboratory solutions (e.g. metal sulfates (such as copper sulfate) and metal nitrates (such as lead nitrate)) must be collected for special disposal rather than emptied down a sink.

Ethics is a set of moral principles by which your actions can be judged as right or wrong. Scientists must obtain approval for their research from an ethics committee and follow ethical guidelines relating to animals and humans. In reality, school science investigations generally have minor, if any, ethical issues. However, consider if one party or group benefits over another during the experiment, or are there real-world connotations of the investigation not accepting the rights of a community group?

Collection of sufficient and relevant raw data

Enough data needs to be collected to substantiate whether a relationship exists between the dependent and independent variables. For the data to be *relevant*, the investigation must collect values for independent and dependent variables as described in the earlier research question section. For the data to be *sufficient*, it must have an appropriate sample size in terms of an appropriate number of replicate trials and an appropriate number of variations of the independent variable. Sample size will impact on the accuracy of the data, the uncertainty within the data collected, and the ability to interpret the data. In general, a minimum of five data points and three measurements per data point (three trials), or three data points with five measurements per data point (five trials) are needed. This provides a sample size consistent with sufficient and relevant data. Consider your expected trend. If you are expecting a logarithmic or another non-linear trend, you will need at least four or five data points – three would not clearly show the trend. Table 1.10.3 below outlines examples of sample size and their effect on the results of an experiment.

TABLE 1.10.3 Examples of sample size and their effect on an experiment

Example of analysed data	Effect of sample size on analysis
	A line graph with two sample points: With only two individual sample points, these results suggest an unsubstantiated relationship between temperature and enzyme activity; that enzyme activity increases in a linear relationship with temperature. It cannot be concluded that the relationship is linear when there are no sample points between temperature, 20 to 30°C.
	Line graph with three sample points using an uneven temperature scale: The spread between the three individual sample points is inappropriate for this experiment as they should be evenly spread. This example produces results with an incorrect relationship between temperature and enzyme activity: it is an artefact of the uneven *x*-axis scale, not a genuine relationship that the enzyme activity seems to increase then slow down as temperature increases.
	Line graph showing limited sample points: The chosen number of individual sample points (3 points) is appropriate. This may be sufficient. However, the next example provides more precision and validity for the relationship.
	Scatterplot with appropriate number and distribution of sample points: The number of individual sample points (5 points) here best demonstrates the relationship between the variables.
	Line graph based on data that had no replicates (only one trial): These results are due to only a single trial for each individual sample point without any replicates. In this example, the measurement at 40°C was a mistake, and without replicates such a mistake may not be identified and will falsely impact the results. Compare this to the graph above with the same variables and averages (of repeat trials) for each of the 5 sample points.

Having determined your sample size, check with your teacher that your protocol, schedule and methodology are appropriate. You need to be able to perform your experiment independently, in the time available in the school laboratory, and with minimal support from your teachers and school laboratory staff.

It is common for experimentation and testing to not go according to plan even if comprehensive background research has been undertaken. Refinements such as reducing or increasing the sample size may be necessary due to timing issues, change to instrumentation or resource limitation. Remember to maintain your scientific journal (logbook) with notes as you go.

If the expected data is not obtained, do not worry. Sometimes a different point of view, such as from a fellow student or a teacher, is all that is needed to find a solution. If the data has been recorded and can be critically and objectively evaluated (limitations of the investigation are identified) in your report, the work is worthwhile. Unexpected data provides students with an opportunity to add depth to their experimental report by critically evaluating issues in the experiment. There may be evidence of extraneous variables (not previously recognised) or confounding factors. Evaluation of the methodology based on evidence collected and suggested improvement(s) is a key section of the scientific report. This is discussed further in Module 1.16.

Record everything in your journal. If you use digital notes, videos and data recording make sure you run a backup. Your dataset may contain both quantitative (values and measurements) and quantitative (observations) data. Note any difficulties encountered or trials which were conducted slightly differently – perhaps a new source of a chemical was provided half-way through, a new measuring instrument used, or a different person determined some readings. This information may be key to your analysis and evaluation. With justification, a data point can be identified as an **anomaly** and removed from the dataset due to being an outlier. This may be due to a mistake or failure to correctly follow the method (See Module 1.6 Anomalies and outliers, also Data processing in Module 1.12). Nevertheless, all values need to be recorded in the raw data table. Choosing not to record certain measurements or observations is invalid, shows bias and is scientifically fraudulent. Quality scientific analysis processes the raw data as it is and is open to any result. Famously, Alexander Fleming discovered penicillin by not discarding an unexpected result.

A workable plan for data collection is for one group member to be responsible for recording. This minimises clutter at the experimental work bench and provides a consistent single data set to share with the group. The recording person should add the measurement uncertainty of each piece of equipment or instrument as a range of data next to the measured value (e.g. measurement ± Δ measurement). Conducting the experiment and collecting data is a group activity; however, all subsequent parts of the task (data processing, analysis, evaluating and concluding) are completed individually. Therefore, it is important that the dataset is shared to the group immediately the experiment finishes. You are advised to not leave the laboratory until you have received your copy of the hard-earned and important dataset, and to always have a backup.

Electronic data acquisition in schools

Electronic data acquisition takes advantage of highly accurate sensors to collect data directly for processing by a computing device (Figure 1.10.5). There are many sensors, probes and instruments available that can measure multiple observations in a single device.

Many smartwatches, fitness trackers and apps in phones include digital sensors and dataloggers that can be accessed and used for some class practicals and experiments (Figure 1.10.6).

Often one of the measurements is time; the user decides what the other measurement (or variable) is by selecting the type of sensor or probe to use. Probes can measure temperature, pH, humidity, UV radiation, gas pressure, conductivity, concentration of gases in air, or the amount of some pollutants in the environment, along with many more phenomena.

FIGURE 1.10.5 Data acquisition software produces real-time graphs that can be downloaded or printed.

FIGURE 1.10.6 Many smartwatches and phones have digital sensors and in-built dataloggers.

Recording is done accurately and can be continuous or manual, and the measurements are saved electronically. The saved measurements can then be accessed via a computer or directly from the tablet screen. The rate at which measurements are recorded is called the sample rate. Data capture rate can be varied from 100 000 times per second to once an hour, and the duration of recording adjusted to a suitable interval (Figure 1.10.7).

FIGURE 1.10.7 Sensors connected to computing devices such as dataloggers, laptops, tablets, smartphones and watches are readily used in the field or lab to measure the concentration of solutions and other key physical measurements. (a) They can be very precise such as the polarimeter being used here to investigate the concentration of sugar solutions. (b) They measure many times per second and more accurately than a human can (c) They are usually easy to use and produce results immediately, for example, providing the temperature of skin upon contact.

Putting it all together

The sample experiment report that follows covers the ISMG criteria for the Finding section about to determining and managing risk. It may be used as a reference tool when writing your own report.

Essential to the Finding section for your report is the risk assessment and collection of sufficient and relevant raw data.

Safety, environmental and ethical considerations

TABLE 2 Identifying and managing risk factors

Hazard	Possible Consequences	Mitigation	Risk Level
Hot water	The skin can be scalded or burnt by the hot water.	Ensure the water bath remains at the temperature set for the experiment. Exercise caution around the water bath.	Low
Slips/ Trips/Falls	Can slip/trip/fall over belongings/equipment if not packed away safely.	Ensure lab safety procedure is followed. Neatly pack away belongings to keep an organised lab environment.	Low
Biological hazard	Milk and lipase could create a smell that is nausea inducing. Milk remnants on equipment, in sink drains and in spills can decompose and breed bacterial contamination.	Exercise caution around the lab. Keep a steady airflow if feeling light-headed. Clean thoroughly after the experiments. Run water down the sink.	Low

Additional considerations that were made:

- wash hands and contact surfaces after handling milk and lipase to avoid exposure to potential biological contaminants
- the correct disposal of environmental waste
- no ethical issues were present.

1.11 Analysing

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- process data using a range of mathematical calculations
- present sample calculations
- analyse data to prepare graph(s) and identify trends
- analyse data to identify precision of evidence (using uncertainty) and accuracy of evidence (using % error).

PROCESSING THE DATA (SAMPLE CALCULATIONS)

Processing the data in numerous ways (including mathematical techniques and graphing) allows any relationships between variables to be made clear. You will analyse trends or patterns in the data, uncertainties and limitations, outliers and results of significance. To prepare your **processed data**, you should consider which of the following mathematical techniques are suitable in your investigation.

The calculations below are explained in Module 1.7 Data Analysis and listed in the Science Inquiry section of the syllabus.

- Mean
- Gradient analysis
- Scatterplot (with best-fit lines, maximum and minimum trendlines and R^2)
- Measurement uncertainty (absolute measurement uncertainty and percentage measurement uncertainty)
- Propagation of measurement uncertainty (addition, subtraction, multiplication, division)
- Uncertainty of the mean (absolute and percentage)

$$\left(\text{Formula: } \Delta\bar{x} = \pm \frac{(x_{max} - x_{min})}{2}\right)$$

$$\left(\text{Formula: percentage uncertainty (\%)} = \frac{\text{absolute uncertainty}}{\text{measurement}} \times \frac{100}{1}\right)$$

- Percentage error

$$\left(\text{Formula: percentage error (\%)} = \frac{\text{measured value} - \text{true value}}{\text{measurement}} \times \frac{100}{1}\right)$$

- Student's *t*-test (p)
- Pearson's correlation coefficient (r)
- Spearman's rank (R^2)

When you have decided which of these calculations are valid for your investigation, prepare a processed data table and include a sample calculation table which shows a single calculation for each. Remember, there are two types of uncertainty – one associated with the measuring equipment (measurement uncertainty) and one associated with the range of trials (uncertainty of the mean). Both values can be expressed in the same units as the value they describe (absolute uncertainty) or converted to a percent of the measurement or mean that they describe (percentage uncertainty).

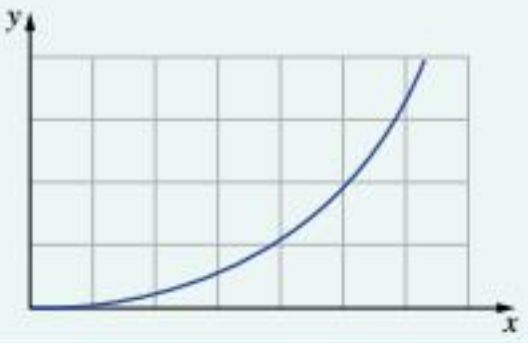
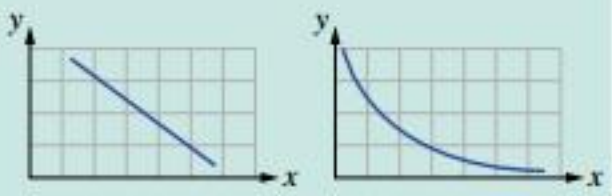
IDENTIFICATION OF TRENDS, PATTERNS AND RELATIONSHIPS

There are a number of ways of presenting processed data, depending on its nature, including tables, graphs, flow charts and diagrams. Usually, a graph is most useful – a clear visual representation showing a direct or inverse relationship between the dependent variable and the independent variable, and if this relationship is linear or not.

Refresh your understanding of graphing by revisiting Module 1.6 Tables and graphing.

Using an Excel program will provide information about the trendline gradient and an R^2 value. By plotting expected values (if known) on the same graph the accuracy of the data is visualised. A number of trends are shown in Table 1.11.1.

TABLE 1.11.1 Graphs showing different trends and relationships between variables

Variables that change in linear or direct proportion to each other produce a straight, sloping trendline. 	Variables that change exponentially in proportion to each other produce a curved trendline. 
When there is an inverse relationship, one variable increases as the other variable decreases. It may be (a) direct or (b) non-linear. 	When there is no relationship between two variables, one variable will not change even if the other changes.

Plotting error bars or maximum and minimum trendlines on a graph indicates if the averages shown are significantly different. Overlapping error bars indicate that the difference between these two values is not statistically significant. The graph(s) that you have prepared provides evidence of the relationship between the dependent and independent variables; however, you should also identify the main features of the graph using a succinct statement. For example:

'As (independent variable) increases (dependent variable) increases/decreases in a linear/non-linear relationship.'

It would be useful to follow this with some specific values from the graph to justify the occurrence of the trend. Further useful analysis could describe the gradient, R^2 value, similarity/difference to the expected values and whether the error bars overlap or not. Remember, the report has an evaluation section so at this stage it is not necessary to fully explain every aspect of the data.

A 'thorough and appropriate identification of trend/patterns/relationships' is required in the Analysis section.

IDENTIFY UNCERTAINTY AND LIMITATIONS OF THE EVIDENCE

Uncertainty, precision and reliability

In science, there is always variation in measurements. Precision and uncertainty were explained in Module 1.5 Measuring precision and accuracy using uncertainties and percentage error. The level of uncertainty is indicated by:

- using values for the measurement uncertainty of the instrument(s) and/or
- using the range of trials and dividing by 2 (uncertainty of the mean).

TABLE 1.11.2 Dataset: volume of 1.0 M hydrochloric acid required to neutralise 25 mL of an unknown concentration of sodium hydroxide solution

Trial	Volume of hydrochloric acid used (mL)
titration 1	20.6±0.05
titration 2	20.9±0.05
titration 3	20.1±0.05
titration 4	20.4±0.05
Mean	20.5±0.4

Each of these can be expressed as percentage of the value they describe, and both give us an indication of the level of random error in the experiment. When identifying and discussing the level of random error, it is most useful to calculate both of these uncertainties, then relate the discussion to the higher value. Keep in mind the general rule that a percentage uncertainty of 5% is the maximum value for a precise and reliable dataset.

Consider these two uncertainty types in an example dataset (Table 1.11.2) which shows the volume of 1.0 M hydrochloric acid required to neutralise 25 mL of an unknown concentration of sodium hydroxide solution.

- The measurement uncertainty due to the precision in the measurement instrument is half a division of the scale, so ± 0.05 mL. Expressed as a percentage of the mean, this is 0.24%
- The uncertainty of the mean is calculated using range (20.9 – 20.1 = 0.8) divided by 2; a result of ± 0.4 mL. Expressed as a percentage of the mean, this is 1.95%.

In this example, the uncertainty of the mean is greater than the measurement uncertainty. So, we can deduce that the precision of the measurement instrument is only slightly contributing to the level of random error in the experiment. We have identified that there is/are further factors contributing to the 1.95% uncertainty of the mean.

In addition, as the 1.95% relative uncertainty of the mean is well below 5%, we can state that the data is precise and reliable, and that the level of random error in the experiment is low.

Percentage error, accuracy and limitations of the evidence

The rationale has provided background information that informs the relationship proposed in the research question. This provides your expected trend and in your analysis you will state if this trend was obtained. In addition, you may be able to research expected values for the data set that you have collected. If this is the case, it is expected that you will calculate percentage error values for each independent variable data point. Module 1.5 explained Measuring precision and accuracy using uncertainty and percentage error with many useful examples.

Percentage error measures how different your data is to expected values and so will allow you to make a judgement about accuracy and validity. Low percentage error indicates low systematic error in the investigation. By researching expected values, you are accessing experimental data from higher-level scientific laboratories. We could expect that the equipment and methodology in a secondary school laboratory is unlikely to accurately replicate these values. For this reason, it is not expected that you use '5%' as a guide for acceptable data (as explained above for uncertainty and precision). Percentage error in a school experiment is likely to be higher than this. Nevertheless, if a percentage error value is as high as 50% you can identify a lack of accuracy, and the investigation is lacking validity. You should not discard the data but identify this as a limitation in your report.

Again, when identifying these issues as part of trends, patterns and relationships, resist the temptation to explain and suggest reasons for these results because this discussion is more relevant to the Concluding and Evaluation sections.

Putting it all together

The sample experiment report that follows, covers the ISMG criteria for Analysing and may be used as a reference tool. Use this as a model for reference when writing your own report.

Results

The Appendix contains the full set of raw data collected.

Table 3: examples of the calculations used to process the raw data.

Table 4: a full processed data set with the calculations from the lipase-milk experiment.

Figure 1: a graph of data from Table 4 showing the mean pH change over 10-minutes for full cream, soy, skim, almond, and lite milk (error bars represented by standard error)

Table 5: A refined processed data table for each milk showing rate of reaction and range.

TABLE 3 Sample calculations showing mean, range, rate of reaction, standard deviation, and standard error

Calculation	Formula	Sample
Average (mean)	$\frac{\text{Trial 1} + \text{Trial 2} + \text{Trial 3}}{3}$	$\frac{7.1\text{pH} + 7.6\text{pH} + 7.2\text{pH}}{3} = 7.3\text{pH}$
Range	starting number − ending number	6.40 pH – 5.83 pH = 0.567 pH
Rate of reaction	$\frac{\text{Range}}{\text{Time taken}}$	$\frac{0.567\text{pH}}{10\text{min}}$ = 0.057 pH
Standard deviation	$\sigma = \sqrt{\frac{\Sigma(x_i - \mu)2}{N}}$ *where σ is standard deviation, x_i is each value from the population, μ is the population mean and N is the size of the population	$\sigma = \sqrt{\frac{(7.1-7.3)2 + (7.6-7.3)2 + (7.2-7.3)2}{3}}$ σ = 0.22
Standard error	$SE = \frac{\sigma}{\sqrt{n}}$ *where SE is standard error, σ is standard deviation, n is the number of samples	$SE = \frac{0.22}{\sqrt{3}}$ SE = 0.13

TABLE 4 Processed data table for the effect of fat concentration in full cream, skim, soy, almond, and lite milk on lipase efficiency (rate of reaction)

Milk	Trial no.	pH of the milk over time (s)										
		0 s	60 s	120 s	180 s	240 s	300 s	360 s	420 s	480 s	540 s	600 s
Full cream	1	6.5	6.5	6.5	6.4	6.4	6.4	6.3	6.2	6.2	6.1	6
	2	6.2	6.2	6.1	6	6	5.9	5.8	5.7	5.7	5.6	5.6
	3	6.5	6.5	6.4	6.3	6.2	6.1	6.1	6	6	5.9	5.9
	Mean pH	6.40	6.40	6.33	6.23	6.20	6.13	6.07	6.0	6.0	5.9	5.8
	Mean rate of reaction	0.00	0.00	0.07	0.10	0.03	0.07	0.07	0.10	0.00	0.10	0.03
	Standard deviation	0.17	0.17	0.21	0.21	0.20	0.25	0.25	0.25	0.25	0.25	0.21
	Standard error	0.10	0.10	0.12	0.12	0.12	0.15	0.15	0.15	0.15	0.15	0.12
Skim	1	6.3	6.2	6.2	6.2	6.2	6.2	6.2	6.2	6.2	6.2	6.2
	2	6.8	6.8	6.8	6.8	6.8	6.8	6.7	6.7	6.7	6.7	6.7
	3	6.6	6.6	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5	6.5
	Mean pH	6.57	6.54	6.50	6.49	6.49	6.48	6.48	6.48	6.48	6.48	6.48
	Mean rate of reaction	0.00	0.04	0.04	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Standard deviation	0.26	0.31	0.30	0.29	0.28	0.28	0.27	0.27	0.27	0.27	0.27
	Standard error	0.15	0.18	0.17	0.16	0.16	0.16	0.16	0.16	0.15	0.15	0.15

Milk	Trial no.	pH of the milk over time (s)										
		0 s	60 s	120 s	180 s	240 s	300 s	360 s	420 s	480 s	540 s	600 s
Soy	1	7.1	7.1	7.1	7.1	7	6.9	6.8	6.8	6.7	6.7	6.6
	2	7.6	7.4	7.3	7.2	7.1	7	7	7	7	6.9	6.9
	3	7.2	7.2	7.1	7	6.9	6.8	6.8	6.8	6.7	6.7	6.7
	Mean pH	7.30	7.23	7.17	7.10	7.00	6.90	6.87	6.87	6.80	6.77	6.73
	Mean rate of reaction	0.000	0.067	0.067	0.067	0.100	0.100	0.033	0.000	0.067	0.033	0.033
	Standard deviation	0.26	0.15	0.12	0.10	0.10	0.10	0.12	0.12	0.17	0.12	0.15
	Standard error	0.15	0.09	0.07	0.06	0.06	0.06	0.07	0.07	0.10	0.07	0.09
Almond	1	7.9	7.9	7.9	7.9	7.8	7.7	7.7	7.7	7.7	7.6	7.6
	2	7.8	7.8	7.8	7.8	7.7	7.7	7.6	7.6	7.6	7.5	7.5
	3*	5.9	6	6	5.9	5.9	5.8	5.8	5.8	5.8	5.8	5.8
	Mean pH	7.20	7.23	7.23	7.20	7.13	7.07	7.03	7.03	7.03	6.97	6.97
	Mean rate of reaction	0.00	-0.03	0.00	0.03	0.07	0.07	0.03	0.00	0.00	0.07	0.00
	Standard deviation*	1.13	1.07	1.07	1.13	1.07	1.10	1.07	1.07	1.07	1.01	1.01
	Standard error	0.65	0.62	0.62	0.65	0.62	0.63	0.62	0.62	0.62	0.58	0.58
Lite	1	6.3	6.2	6.2	6.2	6.2	6.2	6.2	6.2	6.2	6.2	6.2
	2	6.9	6.8	6.7	6.6	6.5	6.5	6.4	6.4	6.4	6.3	6.3
	3	6.4	6.4	6.4	6.3	6.3	6.2	6.2	6.1	6.1	6	6
	Mean pH	6.53	6.47	6.43	6.37	6.33	6.30	6.27	6.23	6.23	6.17	6.17
	Mean rate of reaction	0.00	0.07	0.03	0.07	0.03	0.03	0.03	0.03	0.00	0.07	0.00
	Standard deviation	0.32	0.31	0.25	0.21	0.15	0.17	0.12	0.15	0.15	0.15	0.15
	Standard error	0.19	0.18	0.15	0.12	0.09	0.10	0.07	0.09	0.09	0.09	0.09

*Grey shaded area = outlier, anomalous with respect to the rest of the data.
Mean pH for each milk type is graphed over 10 minutes in Figure 2.

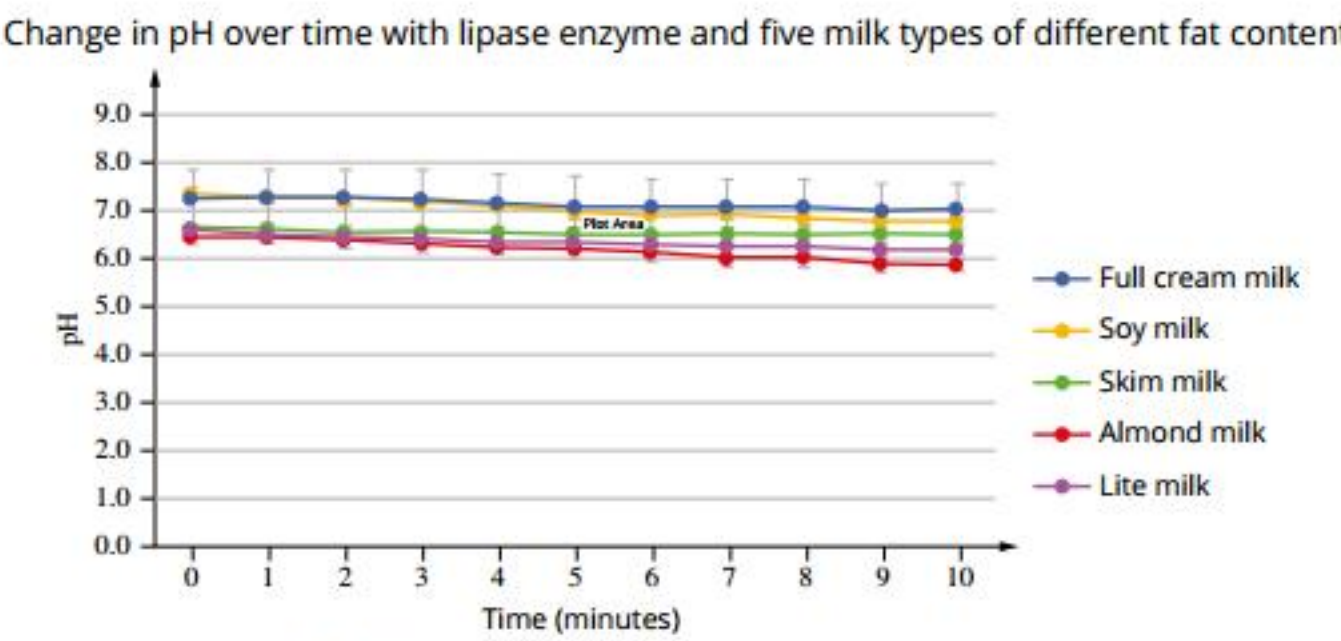


FIGURE 1 Mean pH change over 10-minutes for full cream, soy, skim, almond, and lite milk (error bars represented by standard error)

Interpretation

The data in Table 4 shows the mean pH each milk solution presented after the reaction with lipase present for 10 minutes (600 seconds). The full cream had a starting mean pH of 6.4±0.1 and a final mean pH of 5.8±0.12. The skim milk presented a higher starting pH of 6.57±0.15 and final of 6.48±0.15. The lower standard error of the full cream milk suggests higher precision during data collection compared to the skim milk. Further, the soy and lite milk presented similar results, with soy starting at 7.30±0.15 (lite at 6.53±0.19) and ending at 6.77±0.09 (lite at 6.17±0.09). Both skim, soy, and lite milk have an initial standard error of ±0.15, which decreases as the experiment progresses. A reason for this could be due to initial error or the presence of biological contaminants from past trials. Table 4 also shows that although almond milk started at a pH of 7.2±0.65, at 60 seconds the pH had increased to 7.23±0.62. As the pH increased, this is likely an error with the data collection and not the experiment itself. As such, almond trial 3 was recognised as an outlier.

The graph in Figure 1 presents the mean pH change over 10 minutes for each type of milk in the presence of lipase enzyme.

TABLE 5 Refined processed data table for each milk showing rate of reaction and range

Type of milk	Total monounsaturated fat per 240 mL (grams)	Starting pH	Ending pH	Range	Duration (minutes)	Total rate of reaction (pH/min)
Full cream	8.25	6.40	5.83	0.56	10	0.057
Soy	7.5	7.30	6.73	0.56	10	0.057
Skim	0.25	6.57	6.47	0.09	10	0.010
Almond	4.25	7.20	6.96	0.23	10	0.023
Lite	3.75	6.53	6.16	0.36	10	0.037

Interpretation

The data in Table 5 shows that both the full cream and soy milk had a lipase reaction rate of 0.057 pH/min and were within the same range of 0.56 pH. Although the full cream milk started at 6.4 pH and finished at 5.8 pH, the soy started at 7.3 pH, finished at 6.7 pH, and was within the same range, suggesting that both milks decreased at the same rate. Table 5 also shows the fat content of each milk (per 240 mL), with full cream being the highest (8.25 g), soy being second highest (7.5 g), and skim being the lowest (0.25 g). As expected, skim milk had the slowest rate of reaction (0.01 pH/min) as there was less fat for the lipase to react with. Each trial lasted 10 minutes to ensure a fair test.

Analysis: The research suggests that as the triglycerides are being hydrolysed by the lipase enzyme, the pH of the milks will decrease, as the hydrogen ion concentration increases, thus lowering the pH of the solution (Ahmed, Shah & Ahmed, 2022). The data fits this model for all types of milk used, suggesting all samples reacted in the expected manner. The skim milk data shows a gradual decrease of 0.01 pH/min compared to the almond milk which showed a decrease of 0.023 pH/min. These data points suggest that a lower fat content (lower substrate concentration) reduces the rate of chemical reaction, as there are less collisions that can occur between the active site of the lipase and the substrate. Further, this trend is also supported by the full cream and soy milk which decreased at a rate of 0.057 pH/min and presented the highest fat content. Lite milk also follows these trends. Almond milk was an outlier for Trial 3. As insufficient data was collected for almond milk, to be a fair test, the trials with the error had to be included. As such, although it follows the predicted trends, it is not entirely accurate or precise.

The data generally supports the research model, as full cream has the fastest reaction rate (0.057 pH/min) and is made up of mostly simple saturated fats (~50%), only presenting ~35% monounsaturated and ~15% polyunsaturated. However, soy does not follow this trend as it shares the same rate of reaction, and range, as full cream. Sixty-two per cent of soymilk's fat is polyunsaturated (25% monounsaturated and ~13% saturated) suggesting the rate of reaction should be slower. A reason for this apparent anomaly could be the surface area of the polyunsaturated fats in soymilk, as they have a greater globule surface area exposed to the enzyme's active site (more molecular collisions) (Adair, 2022). Further, soy is closer to the enzyme's optimum pH, allowing for more efficient catalysation. The standard error bars of soy and almond overlap at minutes 0–7 while the other three overlap at minutes 0–4. This suggests the results fall within the same range for all conditions. As the error bars do not overlap outside of these parameters, there is statistical difference between each mean, suggesting that surface area and concentration do influence reaction rate.

1.12 Interpreting and evaluating

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- formulate a conclusion linked to the research question using interpretation of evidence
- justify your conclusion
- evaluate reliability and validity and justify your arguments with evidence
- suggest improvements and extensions.

JUSTIFIED CONCLUSION LINKED TO THE RESEARCH QUESTION

While you have already described the trend observed for the data in the Analysis section, it is important in the conclusion to extend this idea further and relate specifically to the research question. Was the research question supported? Has the research question been fully answered? If not, identify why not. Summarise your findings in a conclusion by writing in an objective and clear manner. State the observed relationship between the dependent and independent variables using the terminology of the research question.

Table 1.12.1 shows examples of concluding statements.

TABLE 1.12.1 Examples of strong and weak conclusions to a research question and hypothesis

	Strong conclusion	Weak conclusion
Research question: How does the temperature affect the pH of water?	Analysis of the results on the effect of an increase in temperature of water from 5°C to 40°C showed an inverse relationship in which the pH of water decreased from 7.4 to 6.7. These results support current knowledge that an increase in water temperature results in a decrease in its pH.	The results show that temperature does affect the pH of water.
Hypothesis: An increase in the temperature of pond water will result in a decrease in the measured pH of the water sample.	An increase in temperature from 5°C to 40°C resulted in a decrease in the pH of the water from 7.4 to 6.7.	The pH of water decreased as temperature increased.

Ways to justify your conclusion include:

- describe the scientific concepts that explain the observed relationship. The key finding should be explained within the right context, meaning the related scientific ideas, concepts and theories. Does the data support the literature or contradict the literature? Within this context, explain the basis for the trend observed
- describe the evidence that led to the data trend. State values that indicate the strength of the relationship between the experimental variables (data, observations or results) including information gained from the errors bars on the graph and the R^2 value(s). Refer to your tables and graphs and cite evidence succinctly
- compare your finding to secondary data you have found, from journal articles or other reliable sources, that is directly connected to the experiment. Explain if the results support your observations. You should report that your results are consistent with the findings. Otherwise, it is essential that you reflect on any contrasting results, and identify or suggest possible explanations why the results differ. This could be due to differences in the measurement technique or other

factors such as the control or measured variables. It might also help you identify possible sources of error within your investigation.

In circumstances where a clear trend is not discernible, you can interpret what the results mean. Interpretations are made when limitations exist related to the methodology employed, the sample size, the instruments used, the variables controlled and the raw data measured. Interpretations are not meant to provide answers and complete explanations, only a valid and reliable plausibility based on what was measured. Interpretations may point the way to future research questions, improvements and directions.

In the conclusion, it is appropriate to briefly describe evidence relating to the quality of experimentation and any key limitations. You have two aspects already identified in Analysis, that is the reliability and the validity (as described by % uncertainty and % error, respectively). Describe which of these was high/low and what this indicates about reliability and validity in your investigation. Resist the temptation to explain these further as this is more usefully done in the Evaluation.

The conclusion is sometimes positioned after the analysis and sometimes at the end of the report. Its placement in your report will need to make sense to you and maintain the flow of ideas:

- Option 1: planning → data collection and analysis → conclusion → evaluation
- Option 2: planning → data collection and analysis → evaluation → conclusion.

JUSTIFIED DISCUSSION OF RELIABILITY AND VALIDITY

Your analysis and conclusion have identified the trend in the data but also identified two other key aspects of the data – reliability and validity. A statement of these has also been made in the conclusion. The syllabus verb in analysis is 'identify' but here, in the Interpreting and Evaluating section, there is 'discuss' for the syllabus descriptor: justified discussion of reliability and validity. This gives you a further opportunity to explain statements made in the analysis related to % uncertainty (reliability) and % error (validity). In your discussion, consider the following:

- Are these low or high?
- What is the effect on the usefulness of the data to answer the research question?
- What could have caused excessive uncertainty and random error?
- What could have caused excessive systematic error?
- Any factors that could affect validity, accuracy, precision and reliability of data.

As you consider this discussion, remember that your data processing has provided extensive evidence – not just the independent-dependent variable relationship but also measurement uncertainty, uncertainty of the mean and percentage error. Use these values to add depth to your discussion of the evidence. For example, if you have calculated the percentage uncertainty associated with the measurement instrument (say, a measuring cylinder) to be 0.24% (see above) then suggesting that the lack of precision of this equipment is the cause of a percentage error of 40% is not reasonable.

Furthermore, if you have calculated the percentage error of the data to range between say 30% to 50% and also show that the percentage error is decreasing during the experiment, your reasoning for the cause of this percentage error needs to relate to a factor which is less at the end of the experiment than at the beginning. An example of this is an enzyme experiment measuring the decomposition of the substrate, hydrogen peroxide. As well as the catabolic reaction catalysed by the enzyme, there will be some spontaneous decomposition of the hydrogen peroxide, as it has some natural instability at room temperature. As a result, we expect the systematic error to increase over the time of the experiment and so percentage error should increase not decrease. This needs to be evident in the data for the syllabus descriptor 'justified discussion of validity' to be met. Both of these are examples of the use of evidence rather than methodology to describe and discuss uncertainty and limitations.

As you discuss factors affecting the quality of the data, remember the definitions of random and systematic error, and describe each factor in terms of one of these. Is the factor expected to always produce a change in the same direction (systematic error, low validity)? Or will the factor produce a more random effect where the value is sometimes higher and sometimes lower (random error, low reliability)? Look carefully at the evidence in your data for each of these. Refresh your understanding of random and systematic error using Figure 1.12.1.

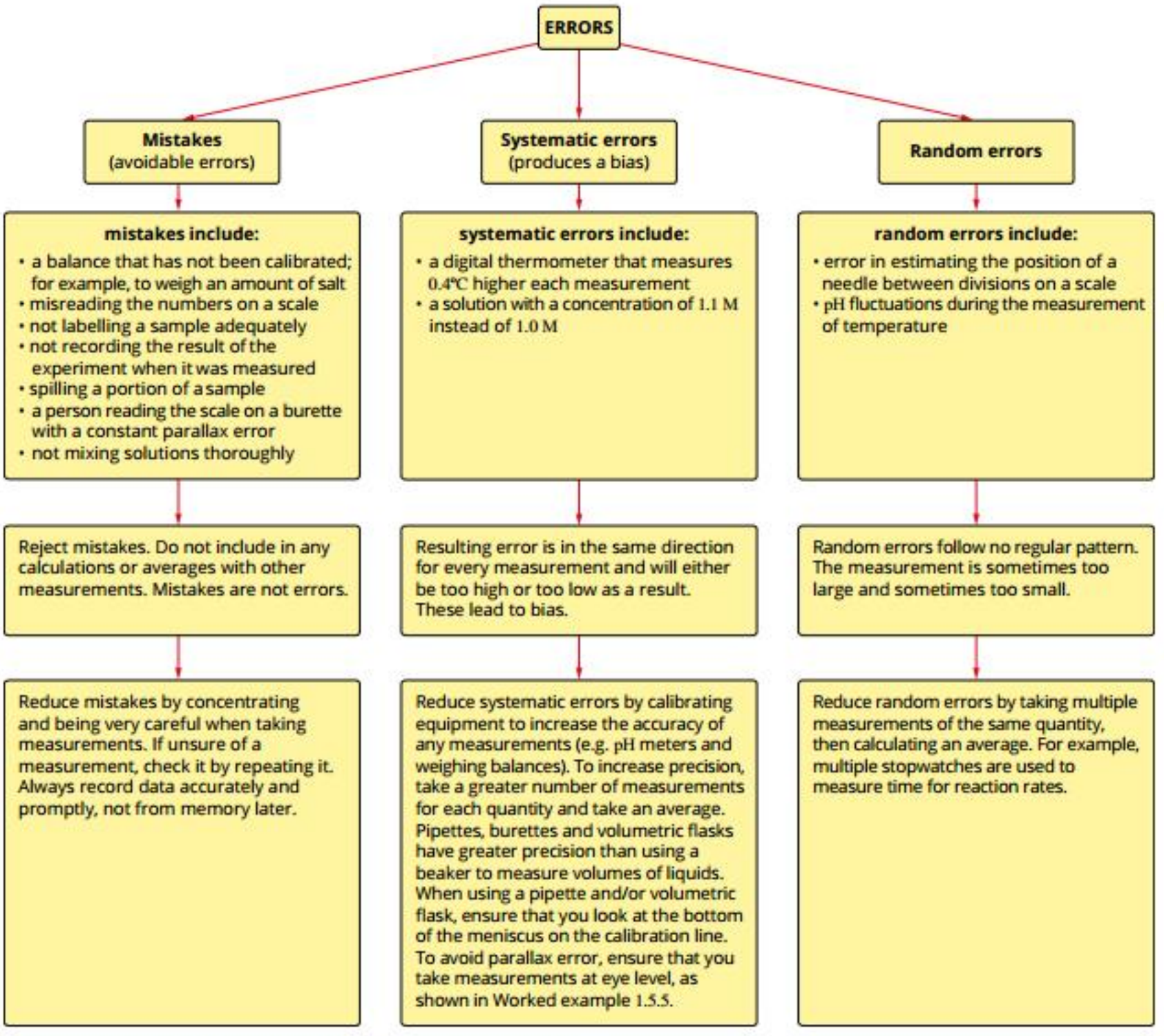

ERRORS
Mistakes
(avoidable errors)
Systematic errors
(produces a bias)
Random errors
mistakes include:
• a balance that has not been calibrated; for example, to weigh an amount of salt
• misreading the numbers on a scale
• not labelling a sample adequately
• not recording the result of the experiment when it was measured
• spilling a portion of a sample
• a person reading the scale on a burette with a constant parallax error
• not mixing solutions thoroughly
systematic errors include:
• a digital thermometer that measures 0.4°C higher each measurement
• a solution with a concentration of 1.1 M instead of 1.0 M
random errors include:
• error in estimating the position of a needle between divisions on a scale
• pH fluctuations during the measurement of temperature
Reject mistakes. Do not include in any calculations or averages with other measurements. Mistakes are not errors.
Resulting error is in the same direction for every measurement and will either be too high or too low as a result. These lead to bias.
Random errors follow no regular pattern. The measurement is sometimes too large and sometimes too small.
Reduce mistakes by concentrating and being very careful when taking measurements. If unsure of a measurement, check it by repeating it. Always record data accurately and promptly, not from memory later.
Reduce systematic errors by calibrating equipment to increase the accuracy of any measurements (e.g. pH meters and weighing balances). To increase precision, take a greater number of measurements for each quantity and take an average. Pipettes, burettes and volumetric flasks have greater precision than using a beaker to measure volumes of liquids. When using a pipette and/or volumetric flask, ensure that you look at the bottom of the meniscus on the calibration line. To avoid parallax error, ensure that you take measurements at eye level, as shown in Worked example 1.5.5.
Reduce random errors by taking multiple measurements of the same quantity, then calculating an average. For example, multiple stopwatches are used to measure time for reaction rates.

FIGURE 1.12.1 Types of errors that can be made in an experiment

Identify any **bias** in any part of the investigation method, including sampling and measurements. Bias is a form of systematic error resulting from the researcher's personal preferences and can include poor definitions, incorrect assumptions, errors in design and methodology. Some biases cannot be eliminated but should at least be addressed in the discussion.

IDENTIFY IMPROVEMENTS AND EXTENSIONS

Once any limitations or problems in the methodology have been identified, recommend improvements for how the investigation could be conducted if repeated. What improvements can be made to aspects of the investigation identified as limitations. What changes to the materials or method would address the lack of reliability or validity of the data? It may not have been possible, due to time or equipment restraints, for you to have made these improvements. It is just required that you identify and suggest them.

Ways to extend the investigation:

- Do the findings lead to further questions?
- Can the findings be extended to another situation?
- What extensions can be investigated for a related scientific idea?
- Do the experimental findings have broader or further practical implications?

In the modifications section, you provided a reason for each change that you made to the original experiment. In a similar way provide a reason for each improvement and extension. Show a logical relationship between limitations of the evidence or ways the research question has not been addressed and future experimentation. Statements such as 'test more substances' or 'complete more trials' do not match a high-level response ('logically derived' ISMG descriptor) and will better match lower descriptors ('related' or 'ineffective').

Figure 1.12.2 provides some guiding questions for this process.

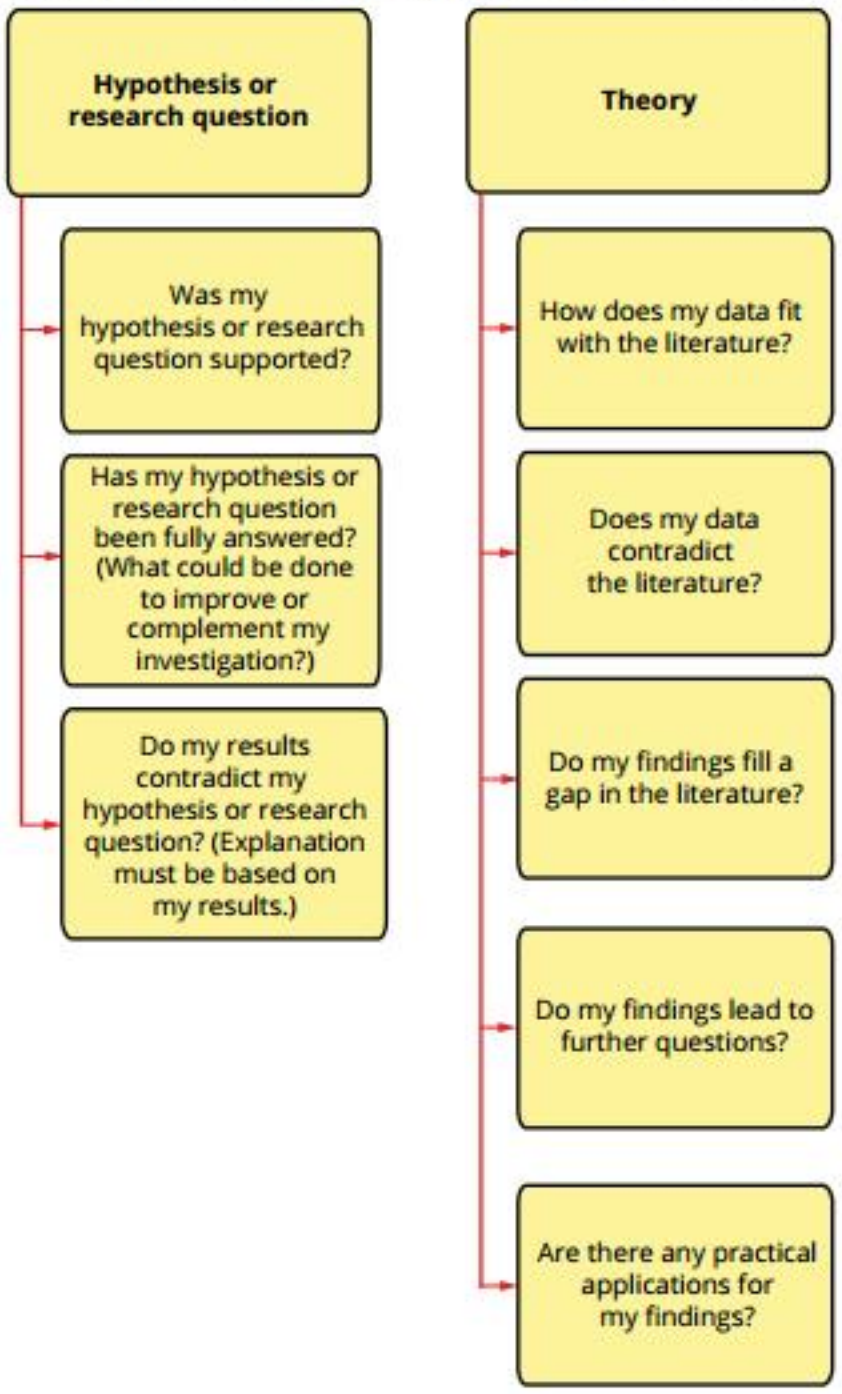

Hypothesis or research question
Was my hypothesis or research question supported?
Has my hypothesis or research question been fully answered? (What could be done to improve or complement my investigation?)
Do my results contradict my hypothesis or research question? (Explanation must be based on my results.)
Theory
How does my data fit with the literature?
Does my data contradict the literature?
Do my findings fill a gap in the literature?
Do my findings lead to further questions?
Are there any practical applications for my findings?

FIGURE 1.13.2 Use these questions to help identify improvements and extensions to further answer the research question and the associated theory.

The syllabus descriptor states, 'logically derived' and so it is important to limit your suggestions for improvements and extensions to experimental issues that have already been identified (in analysis and evaluation). Conversely, if you have described a key limitation of the experiment related to high levels of random or systematic error, you should describe improvement(s) for this issue.

Interpreting and evaluating: Putting it all together (justified conclusion, justified discussion of reliability and validity, improvements and extensions)

Putting it all together

The sample experiment report that follows covers the ISMG criteria for the Interpreting and evaluating section. Use this as a model for reference when writing your own report.

Effect of lipid and triglyceride concentration on lingual lipase efficiency in full cream, almond, soy, lite and skim milk.

Conclusion

An in-depth experiment was conducted to answer the research question: How does changing the lipid concentration in milks (full cream, lite, soy, almond, and skim) impact the efficiency (rate of reaction) of lingual lipase enzyme when tested at 37°C, over a 10-minute period?

Lucas (2020) had indicated that by increasing the concentration of the substrate lipids and triglycerides present in the milk solutions, the rate of chemical reaction with lipase would also increase. This idea was supported by the results collected through the experiment, suggesting that 240 mL of full cream milk, at a fat concentration of 8.25 g/240 mL, would have a mean reaction rate of 0.057 pH/min when reacting with 5 mL of lingual lipase, and presented the fastest rate of reaction (equal with soy milk). Furthermore, the limitations of the experiment, namely the fluctuation of temperature noted below, provided the results with marginal error and lack of precision. However, as they were continuous throughout, the effect on the data was considered negligible. In future trials, it is recommended that a larger sample size of milk types is investigated and possibly exploring pancreatic and gastric lipase enzymes to further corroborate and compare the findings. In addition, it was evident that skim milk with complex triglyceride chains and the lowest fat content at 0.25 g/240 mL had the slowest mean reaction rate, recording only 0.01 pH/min.

Evaluation

Limitations and improvements

The experiment produced data that was conclusive and deemed reliable. However, as can be expected for school-based research, there were limitations to the experiment.

A limitation that was present and could not be prevented, was the pH meter that was used presented an absolute uncertainty of ±0.01 pH. However, due to the marginal difference this limitation presented, the difference was negligible. In future trials, a recommended improvement would be to use a SevenExcellence pH meter s400, which is accurate to ±0.001 pH. The greater precision of the device would allow for a more thorough and accurate collection of data, from which to draw more specific conclusions.

An improvement for equipment would be to access a hotplate magnetic stirrer for the milk-lipase liquid to be heated and mixed uniformly at the same time. This would maintain a homogenous mix for pH measurement at the sensor position (Figure 2 in Appendix).

Further, another limitation to the experiment was the possibility for biological contaminants, identified as likely in the almond milk trials, where the standard deviation presented >1.00. As the solutions used in the experiment were not biologically screened prior to use, it cannot be confirmed or denied that a biological catalyst/contaminant other than lingual lipase may have been present. Research indicated that a biological contaminant (if present) could skew data greatly, possibly increasing the toxicity of the solution (due to cell apoptosis), therefore reducing the validity of the results. To improve this limitation, it was recommended that the solutions be screened for purity to ensure that the data incorporates this possibility.

Another substantial limitation present was that the temperature of the water bath fluctuated beneath the optimum 37°C periodically. Although the point at which the temperature fluctuated and the degree it fluctuated was not recorded, it was noted that it occurred throughout the entire experiment. Depending on the severity of the temperature variation, the enzymes may have been energised or de-energised, and as such influence the validity of the results. However, this limitation was considered negligible as it occurred throughout all trials, keeping the test fair despite providing some error. In future trials, it was recommended that the beakers remained in water at constant temperature to reduce the likelihood of error, with a prediction this would improve the results.

In future trials an extension is suggested. To better investigate how the concentration of lipid substrate impacts reaction rate, it is recommended that the sample size is increased. In doing so, a greater range of data could be collected as well as provide more accurate results as to how different lipid and triglyceride types influence lingual lipase efficiency. It was also posed that perhaps other types of lipases should be investigated. By doing so, the accuracy of the results may not change, yet a greater understanding can be reached.

Sources of Error

Affecting reliability

The temperature of the solution was not maintained precisely after being heated to 37°C. Therefore, it is not known reliably what specific temperature in each trial was affecting the rate of reaction with the lipase and milk lipids.

The samples were not randomly selected, and the exact amount of lipase was not consistent thus meaning random variation exists between the samples. This could explain some of the imprecision within the data.

Each test tube was not thoroughly cleaned prior to use. Therefore, it is not known if other biological factors may have affected the rate of reaction.

The electronic device used to measure pH was precise to ±0.01, thus contributing to some imprecision of the data. However, it was evident other factors contributed more to the data's imprecision and as such the inaccuracy was not analysed for significance.

Affecting validity

The pH device recorded the pH of the solution closest to the sensor, therefore if the lipase was not mixed evenly throughout the solution, the pH sensor would not process all changes of pH. This could contribute to inaccuracy in the data.

As the milk used was not screened prior to the test, potential biological contaminants could exist. As such, this could contribute to reducing the validity and precision of the results.

Temperature fluctuations present may skew and alter the outcome of the trials. It was considered yet deemed negligible as the test remained fair.

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APPENDIX

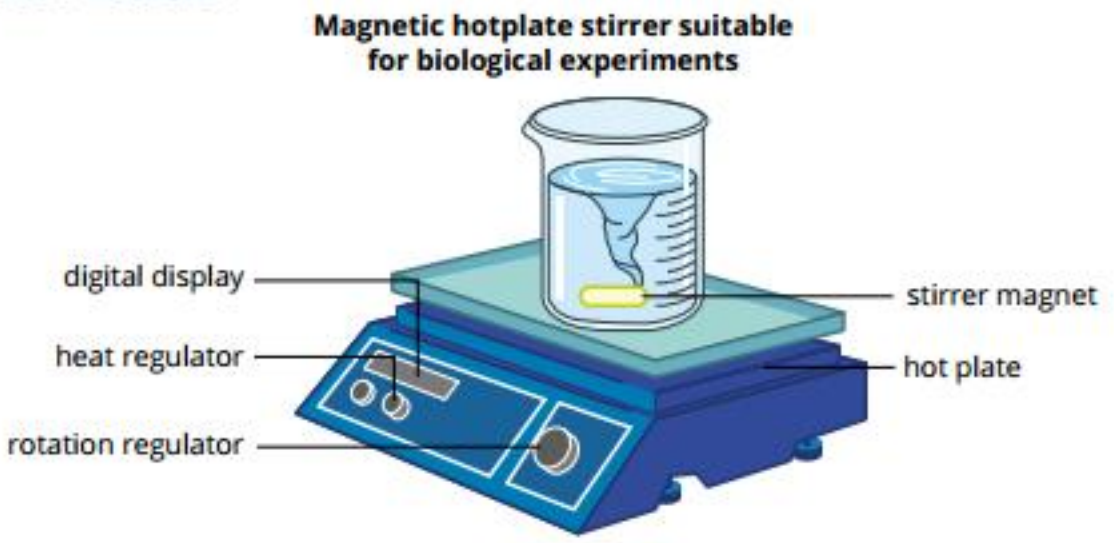

FIGURE 2 Equipment suitable to improve the lingual lipase experiment

TABLE 6

Time (minutes)	Full cream milk				Rate of reaction (pH/min)	Standard deviation	standard error
	Trial 1 (pH)	Trial 2 (pH)	Trial 3 (pH)	mean (pH)			
0	6.5	6.2	6.5	6.4	0.000	0.17	0.10
1	6.5	6.2	6.5	6.4	0.000	0.17	0.10
2	6.5	6.1	6.4	6.3	0.067	0.21	0.12
3	6.4	6	6.3	6.2	0.100	0.21	0.12
4	6.4	6	6.2	6.2	0.033	0.20	0.12
5	6.4	5.9	6.1	6.1	0.067	0.25	0.15
6	6.3	5.8	6.1	6.1	0.067	0.25	0.15
7	6.2	5.7	6	6.0	0.100	0.25	0.15
8	6.2	5.7	6	6.0	0.000	0.25	0.15
9	6.1	5.6	5.9	5.9	0.100	0.25	0.15
10	6	5.6	5.9	5.8	0.033	0.21	0.12

Time (minutes)	Skim milk				Rate of reaction (pH/min)	Standard deviation	standard error
	Trial 1 (pH)	Trial 2 (pH)	Trial 3 (pH)	mean (pH)			
0	6.3	6.8	6.6	6.57	0.000	0.26	0.15
1	6.2	6.8	6.6	6.54	0.037	0.31	0.18
2	6.2	6.8	6.5	6.50	0.040	0.30	0.17
3	6.2	6.8	6.5	6.49	0.007	0.29	0.16
4	6.2	6.8	6.5	6.49	0.003	0.28	0.16
5	6.2	6.8	6.5	6.48	0.003	0.28	0.16
6	6.2	6.7	6.5	6.48	0.003	0.27	0.16
7	6.2	6.7	6.5	6.48	0.000	0.27	0.16
8	6.2	6.7	6.5	6.48	0.003	0.27	0.15
9	6.2	6.7	6.5	6.48	0.000	0.27	0.15
10	6.2	6.7	6.5	6.48	0.000	0.27	0.15

Time (minutes)	Lite milk				Rate of reaction (pH/min)	Standard deviation	standard error
	Trial 1 (pH)	Trial 2 (pH)	Trial 3 (pH)	mean (pH)			
0	6.3	6.9	6.4	6.53	0.000	0.32	0.19
1	6.2	6.8	6.4	6.47	0.067	0.31	0.18
2	6.2	6.7	6.4	6.43	0.033	0.25	0.15
3	6.2	6.6	6.3	6.37	0.067	0.21	0.12
4	6.2	6.5	6.3	6.33	0.033	0.15	0.09
5	6.2	6.5	6.2	6.30	0.033	0.17	0.10
6	6.2	6.4	6.2	6.27	0.033	0.12	0.07
7	6.2	6.4	6.1	6.23	0.033	0.15	0.09
8	6.2	6.4	6.1	6.23	0.000	0.15	0.09
9	6.2	6.3	6	6.17	0.067	0.15	0.09
10	6.2	6.3	6	6.17	0.000	0.15	0.09

Time (minutes)	Soy milk				Rate of reaction (pH/min)	Standard deviation	standard error
	Trial 1 (pH)	Trial 2 (pH)	Trial 3 (pH)	mean (pH)			
0	7.1	7.6	7.2	7.30	0.000	0.26	0.15
1	7.1	7.4	7.2	7.23	0.067	0.15	0.09
2	7.1	7.3	7.1	7.17	0.067	0.12	0.07
3	7.1	7.2	7	7.10	0.067	0.10	0.06
4	7	7.1	6.9	7.00	0.100	0.10	0.06
5	6.9	7	6.8	6.90	0.100	0.10	0.06
6	6.8	7	6.8	6.87	0.033	0.12	0.07
7	6.8	7	6.8	6.87	0.000	0.12	0.07
8	6.7	7	6.7	6.80	0.067	0.17	0.10
9	6.7	6.9	6.7	6.77	0.033	0.12	0.07
10	6.6	6.9	6.7	6.73	0.033	0.15	0.09

Time (minutes)	Almond milk				Rate of reaction (pH/ min)	Standard deviation	standard error
	Trial 1 (pH)	Trial 2 (pH)	Trial 3 (pH)	mean (pH)			
0	7.9	7.8	5.9	7.20	0.000	1.13	0.65
1	7.9	7.8	6	7.23	-0.033	1.07	0.62
2	7.9	7.8	6	7.23	0.000	1.07	0.62
3	7.9	7.8	5.9	7.20	0.033	1.13	0.65
4	7.8	7.7	5.9	7.13	0.067	1.07	0.62
5	7.7	7.7	5.8	7.07	0.067	1.10	0.63
6	7.7	7.6	5.8	7.03	0.033	1.07	0.62
7	7.7	7.6	5.8	7.03	0.000	1.07	0.62
8	7.7	7.6	5.8	7.03	0.000	1.07	0.62
9	7.6	7.5	5.8	6.97	0.067	1.01	0.58
10	7.6	7.5	5.8	6.97	0.000	1.01	0.58

PART C RESEARCH INVESTIGATION

The research investigation assessment (IA3) requires students to investigate a claim, provided by their teacher, by drawing on secondary evidence from scientific texts. Students will make use of this research to analyse, interpret and evaluate the evidence to reach a justifiable conclusion about the research question. The task requires students to locate and use information beyond the scope of their knowledge and the data they have been given.

QCAA Syllabus states that students must work individually to develop and investigate their own research question. It is recommended that students develop a response in approximately 10 hours of class time and/or their own time.

The research investigation constitutes 20% of the total subject score and may be presented in:

- written form of up to 2000 words, or
- multimodal form using at least two modes delivered simultaneously and up to 11 minutes.

INSTRUMENT-SPECIFIC MARKING GUIDE

As with the student experiment, responses are assessed against an instrument-specific marking guide (ISMG). In developing the research report, it is important to keep in mind the assessment objectives, particularly the characteristics that are described in the performance level descriptors.

- The ISMG has four criteria. Each criterion carries equal marks:
  - forming and finding
  - analysing
  - interpreting
  - evaluating.

Each criterion has performance levels, against which the qualities of the response are assessed. Table C below shows the high-performance level characteristics for the four criteria.

- A performance level is comprised of a performance level descriptor and mark (or mark range).
- The performance level descriptor describes the characteristics that are demonstrated by a response of that quality.

Table 1.14.1 shows the ISMG criteria and elaborations for the highest performances of the research investigation for Unit 4.

Note that in Year 11, this internal assessment task will align with Unit 2 'Maintaining the internal environment' and not Unit 4 'Heredity and continuity of life'. A research project based on IA3, but undertaken in Year 11, will require modification of assessment objectives to relate to Unit 2 subject matter. The ISMG marking guide descriptors and module references remain the same.

TABLE C The ISMG criteria and highest performance descriptors for marking the research report

Criteria	Assessment objectives	ISMG descriptors – Criteria are demonstrated by	Marks	Module
Forming and finding	• Describe ideas and findings about DNA, genes and the continuity of life on Earth. • Apply understanding of DNA, genes and the continuity of life on Earth to develop research questions. • Investigate phenomena associated with DNA, genes and the continuity of life on Earth through research.	• considered rationale identifying clear development of the research question from the claim • a specific and relevant research question • selection of sufficient and relevant sources • appropriate use of genre conventions • acknowledgment of sources of information through appropriate use of referencing conventions	5	1.15 1.8
Analysing	• Analyse research data about DNA, genes and the continuity of life on Earth.	• the identification of sufficient and relevant evidence • thorough identification of relevant trends, patterns and relationships in evidence • thorough and appropriate identification of limitations of evidence	5	1.16
Interpreting	• Describe ideas and findings about DNA, genes and the continuity of life on Earth. • Interpret research evidence about DNA, genes and the continuity of life on Earth.	• justified scientific argument/s • justified conclusion linked to the research question • fluent and concise use of scientific language/ representations	5	1.17 1.8
Evaluating	• Evaluate research processes, claims and conclusions about DNA, genes and the continuity of life on Earth.	• justified discussion of the quality of evidence • extrapolation of credible findings of the research to the claim • suggested improvements and extensions to the investigation that are considered and relevant to the claim	5	1.18

In Part C Modules 1.15 to 1.18, each stage of the development of research investigation will be discussed and aligned with the relevant ISMG criteria, and elaborations for the highest performances.

The research question should specifically address one of the concepts associated with the claim. Commence by identifying a claim and developing your own research question based on that claim. Since this assessment relies heavily on access to research, students need to be able to easily find sources relevant to the research question. Throughout the process, be mindful that scientific language and representations are used (Refer to Module 1.8 Scientific writing).

The final report for the student experiment may be organised under the four criteria headings in the instrument-specific marking guide (ISMG):

- forming and finding
- analysing
- interpreting
- evaluating

However, this is not mandated, and you may elect to organise your final report in a different but logical order using alternate headings. You should start by using the four headings to meet all criteria. Later, you may decide to change the structure to one that is more fluent and concise. Remember the limit for a written report is 2000 words, and the time limit for a multimodal presentation is up to 11 minutes.

Note that items in the Appendix are not included in the word count and are not used for determining levels of achievement.

1.13 Forming and finding

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand and select a claim
- classify elements of the claim
- unpack the elements to identify variables, terms and concepts related to the claim
- plan research and locate sources relevant to the claim
- distinguish between primary and secondary sources
- locate a range of primary and secondary sources
- use a scientific journal to record the information in the sources
- refine the concepts to develop an investigable research question
- understand the criteria against which the research investigation will be assessed.

The components of the Finding and forming criterion are outlined in Module 1.14, Table 1.14.1.

To achieve the highest mark (5) for Forming and finding, you need to:

- Create a research question clearly linked to the subject matter that identifies measurable variables (independent and dependent), as well as a control variable specific to the research question.
- Construct a rationale that clearly outlines how the research question is developed from the original claim. A research rationale outlines the reason to conduct research on your chosen claim i.e. it is the justification for the investigation. This is achieved by conducting a thorough literature review to understand what is already known in the chosen topic and to identify the syllabus concepts that are related to the topic.
- Be aware that the structure and style of the report, quality of sources and use of referencing are also assessed.

UNDERSTANDING THE NATURE OF A CLAIM

A claim is an assertion made without any accompanying evidence to support it. The claim can be a sentence, statement within a sentence, title of an article, quote or anything published in any form. The claim should be in isolation from any justification. The claim is a broad statement that provides access to a range of topics without data, research, evidence or reasoning from known information to support it.

Selecting a claim

If you are provided with a choice of claims, you should ask yourself the following questions about the claims.

Which claim(s):

- interest me or are most relevant to me?
- will have secondary evidence available to me?
- are easy to break down and link to the subject matter in the syllabus unit?
- are broad enough to allow me to undertake a unique investigation?

The answers to these questions will help you decide on the most appropriate claim for you.

To develop the research question from a claim, students must analyse, understand and identify one or more of the following:

- known scientific concepts
- variables
- measurable terms
- ideas related to concepts
- a term that is stated to influence another.

Record the information you collect, your thoughts and analysis in a hard copy or electronic journal. When you write the report, this information will be useful in writing the rationale that documents how the research question was developed from the claim. Write down all the elements found in the claim and try to categorise them using the above list. Each element within the claim may suit more than one category. Table 1.13.1 examples of analysing and categorising elements of a claim.

TABLE 1.13.1 Three examples of analysing and classifying elements of a claim

Example claim 1: Plastics are bad for the environment		
Elements	**Classifying elements of the claim**	**Unpacking the elements**
Plastics are made from crude oil. Define the term 'bad'.	a term that is stated to influence another	hydrocarbons
	a term that is stated to influence another	polymers
	ideas that relate to concepts	non-renewable resource
	measurable terms variables	manufacture requires quantifiable resources and energy damages the environment
Plastics are disposed of and not recycled.	known scientific concepts	most plastics can be recycled so not all are waste
Plastics release toxic chemicals.	ideas that relate to concepts	breakdown in environment
	known scientific concepts	chemicals released from combustion additives
Plastics are not biodegradable.	measurable terms	polymers are inert
	ideas related to concepts	not easily broken down in nature by air, water and sunlight

Example claim 2: Natural shampoos are better than commercial shampoos		
Elements	**Classifying elements of the claim**	**Unpacking the elements**
Define the term 'better'.	measurable terms variables	more environmentally friendly
	ideas related to concepts variables	improved cleaning efficiency
Natural shampoos do not contain harmful chemicals.	a term that is stated to influence another	synthetic chemicals are toxic
Natural shampoos are as effective cleaning products as commercial shampoos.	known scientific concepts	how surfactants work
	measurable terms known scientific concepts	cleaning effectiveness is measurable and related to surface tension, wetting time, solid content, and foaming ability

Example claim 3: Green chemistry is clean chemistry		
Elements	**Classifying elements of the claim**	**Unpacking the elements**
Green chemistry	a term that is stated to influence another	green solvents
	ideas related to concepts	preventing pollution
	ideas related to concepts	renewable energy
	a term that is stated to influence another measurable terms variables	atom economy—higher percentage of useful products formed in the reaction
	a term that is stated to influence another	renewable materials
Environment	measurable terms variables	fewer pollutants released
	measurable terms variables	fewer resources required

Elements	Classifying elements of the claim	Unpacking the elements
Clean chemistry	ideas related to concepts	non-toxic
	measurable terms variables	less energy required
	measurable terms	reduced reagents

Locating sources

It is important to recognise that this is not a university quality report as students do not have the time, access or understanding to analyse the information provided in the scientific research journals. In addition, the research investigation is limited to 2000 words in written form or up to 11 minutes in multimodal form, so does not allow for the depth of investigation of a research journal.

When gathering scientific evidence for the research investigation, source it from reputable publications including:

- scientific journals: research papers and scientific reviews
- scientific articles written by organisations who apply scientific research to their industry
- commercial articles such as science magazines, newspapers and websites.

The research investigation must include a reference list of sufficient and relevant sources. **Sufficient** is defined as 'enough or adequate for the purpose', and **relevant** as 'bearing upon or connected with the matter in hand; to the purpose; applicable and pertinent; has direct bearing on'. Figure 1.13.1 points out the features of sources deemed sufficient and relevant.

Refer back to Module 1.5 to recall the meanings of and distinctions between; valid, reliable, accurate, precise. The glossary of your Student Book is also a valuable resource to quickly check word definitions.

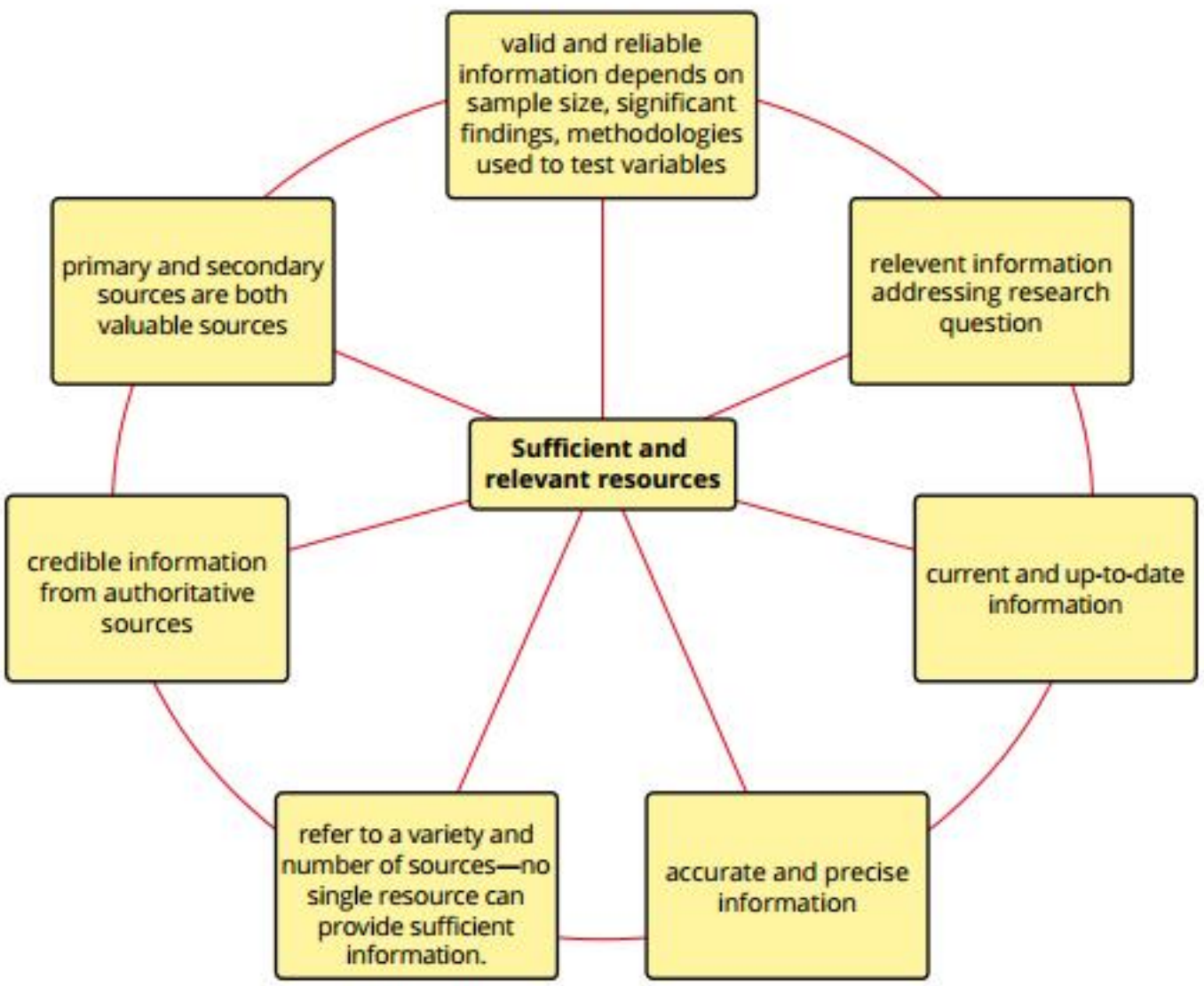
valid and reliable information depends on sample size, significant findings, methodologies used to test variables
primary and secondary sources are both valuable sources
relevent information addressing research question
Sufficient and relevant resources
credible information from authoritative sources
current and up-to-date information
refer to a variety and number of sources—no single resource can provide sufficient information.
accurate and precise information

FIGURE 1.13.1 Features of sources suitable for the research investigation

A good place to start research is to look for interesting sites and textbooks to identify relevant keywords and concepts rather than immediately searching for complex research articles. This could include videos, general scientific articles, even Wikipedia, to gain a general understanding of the topic, or possibly an AI source but only after reviewing Module 1.8 Using AI in research and writing. Be cautious with content in user-generated platforms as they usually contain bias and inaccuracies.

Access available open-access sources; some of which can be identified using educational, government or industry websites, with suffixes including .edu, .gov or .org. There are academic search engines such Google Scholar and Semantic Scholar to find peer reviewed articles. Be aware that both search engines use AI to search for your requested articles and that may involve some bias. However, this does not mean the article itself is AI-generated.

Where necessary, you can often freely access scientific journals through academic networks, such as ResearchGate, Academic.edu and ScienceDirect.com. They will narrow your research to the relevant topic with some, but not all, open access articles. If the article has not already been published, you can request a copy of the article from the author or ask them a question. Your school librarian can help here.

When accessing the sources, start with the title which should include some of the keywords related to the concept. As you refine the focus of your investigation, use the evidence to refine the claim and develop the rationale. Your search will narrow and lead to data tables, graphs, images and abstracts. If the abstract is useful, download the article or request a copy of it. Look at the images with their titles, and the conclusion. These may be more useful than the text of the article.

Remember to document the sources and summarise the information contained in your journal to facilitate the writing of the report and generate a reference list. Consider whether the information you use is from a primary or secondary source. Some sources provide help with the citation of scientific articles. APA is the recommended referencing style.

A useful way to collate the evidence is in a table with headings such as:

- Article overview—summarises the article
- Evidence—outlines the methodology, trends, patterns and relationships reported in the study, and limitations
- Conclusions—summarises the evidence
- Relationship to research question—includes your thoughts and highlights the parts of the research question that this evidence addresses
- Limitations—identifies aspects of the research question that are not addressed or cannot be answered due to the methodology
- Source—identifies primary or secondary citation.

Primary and secondary sources

Table 1.13.2 summarises the characteristics of primary and secondary sources. Sometimes the same information may be contained within a primary and a secondary source, depending on when and by whom it was written. For example, a scientist's journal article on a clinical trial of treatments for teenage obesity is a **primary source**, while a general magazine article about teenage obesity, written by a journalist and referring to the scientific study, is a **secondary source**.

TABLE 1.13.2 Summary of primary and secondary sources

	Primary sources	Secondary sources
characteristics	• first-hand records of events or experiences • written at the time the event happened • original documents	• interpretations of primary sources • written by people who did not see or experience the event • use information from original documents but rework it
examples	• results of experiments • scientific journal/magazine articles by the original researchers • reports of scientific discoveries • photographs, specimens, maps and artefacts • interviews with experts • websites (if they meet the criteria above)	• textbooks • biographies • newspaper articles • magazine articles • radio and television documentaries • websites that interpret the scientific work of others • podcasts

A primary source is written by the observer/witness of an event or the scientist who conducted the research. The information has only been processed by the original observer; therefore it is the least biased of all available sources of information. However, even primary sources may be biased, as the observer or researcher had to make choices related to the observation, control of variables, use of instruments and choices for processing data.

Secondary sources of information are not eyewitness accounts but interpretations of events by other people. As second-hand information, the accuracy and reliability may be reduced, events may be interpreted through the writer's perception and bias, there may be omissions, or material taken out of context. You should aim to use a wide range of data sources when using secondary data, to cross-check for accuracy, reliability and validity of information. A single source will not be accepted as 'sufficient'. Even with multiple sources, the same information may have been repeatedly copied from a single secondary source, rather than the original primary source.

When searching for information and evidence, follow these guidelines. You could use a checklist.

- Determine if it is a primary or secondary resource.
- Confirm that it is valid.
  - Check that it contains information that is specifically related to the claim.
  - Check the evidence and information is pertinent to the variables in the research question.
- Assess its reliability.
  - Is it current/recent information?
  - Is it up-to-date in its understanding of relationships?
  - Is the evidence equivalent to other sources?
  - Check credibility—consider who the author is, their qualifications and expertise.
  - Evaluate the methodology, including what variables were controlled or measured.

Articles in scientific journals

Peer-reviewed scientific journals are excellent sources of information. They are usually focused on one area of science, with a collection of scientific reports written by scientists who conducted research. The reports and articles found in scientific journals are published primary sources, meaning they are the results of the experiments (Figure 1.13.2).

OPEN ACCESS Freely available online

PLOS ONE

Squeezing the Muscle: Compression Clothing and Muscle Metabolism during Recovery from High Intensity Exercise

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Abstract

The purpose of this experiment was to investigate skeletal muscle blood flow and glucose uptake in m. biceps (BF) and m. quadriceps femoris (QF) 1) during recovery from high intensity cycle exercise, and 2) while wearing a compression short applying ~37 mmHg to the thigh muscles. Blood flow and glucose uptake were measured in the compressed and non-compressed leg of 6 healthy men by using positron emission tomography. At baseline blood flow in QF ($P=0.79$) and BF ($P=0.90$) did not differ between the compressed and the non-compressed leg. During recovery muscle blood flow was higher compared to baseline in both compressed ($P<0.01$) and non-compressed QF ($P<0.001$) but not in compressed ($P=0.41$) and non-compressed BF ($P=0.05$; effect size = 2.74). During recovery blood flow was lower in compressed QF ($P<0.01$) but not in BF ($P=0.26$) compared to the non-compressed muscles. During baseline and recovery no differences in blood flow were detected between the superficial and deep parts of QF in both, compressed (baseline $P=0.79$; recovery $P=0.68$) and non-compressed leg (baseline $P=0.64$; recovery $P=0.06$). During recovery glucose uptake was higher in QF compared to BF in both conditions ($P<0.01$) with no difference between the compressed and non-compressed thigh. Glucose uptake was higher in the deep compared to the superficial parts of QF (compression leg $P=0.02$). These results demonstrate that wearing compression shorts with ~37 mmHg of external pressure reduces blood flow both in the deep and superficial regions of muscle tissue during recovery from high intensity exercise but does not affect glucose uptake in BF and QF.

Citation: Sperlich B, Born D-P, Kaskinoro K, Kalliokoski KK, Laaksonen MS (2013) Squeezing the Muscle: Compression Clothing and Muscle Metabolism during Recovery from High Intensity Exercise. PLoS ONE 8(4): e60923. doi:10.1371/journal.pone.0060923

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Competing Interests: BS and DPB had travel costs to Turku, Finland, paid by SIGVARIS for data acquisition. BS has received consultancy fee by SIGVARIS on other previous projects other than the present one. This does not alter the authors' adherence to all the PLOS ONE policies on sharing data and materials. None of the other authors have any ownership of stocks, employment or board membership at SIGVARIS AG or other companies with competing interests with relation to data presented here, neither financial, professional, nor personal.

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Introduction

Skeletal muscle blood flow incorporates a key role in aerobic muscle metabolism matching the delivery of oxygen and energy substrates for energetic demands, as well as the transportation of waste products and heat from the muscle tissue. The response of skeletal muscle blood flow and metabolism at the onset of and during exercise are well documented. In general, muscle blood flow increases rapidly with an exercise-depended plateau after approximately 30 s of exercise [1,2]. Muscle blood flow is heterogeneously distributed [3] in a manner that it is higher in the deeper compared to the superficial parts of the m. quadriceps femoris [4].

Another point is that blood glucose concentration plays an important role in restoring muscle glycogen during recovery from exercise [5]. Unfortunately, little is known about the crucial role of blood flow and its association to glucose uptake, during recovery from high intensity exercise [6] and so far no study investigated this matter in connection with the application of compression clothing.

In the past two decades, various forms of compression clothing have been applied by elite and recreational athletes due to accumulating evidence regarding the possible performance [7,8] and recovery [9–11] enhancing properties. Early research showed that the application of 15 mmHg external pressure while lying in supine position reduces the cross-sectional area of the venous systems (from 2.65 cm^2 to 0.53 cm^2) thereby elevating mean linear blood flow velocity (from 0.5 cm/s to 2.5 cm/s) [12] and thus reducing contact time and thrombogenesis [13]. The application of compression seems to enhance muscle blood flow [14] and has been associated with enhanced clearance of metabolites, such as blood lactate [15,16]. However, it also has been suggested that reduced levels of blood lactate are attributable to a greater retention of the molecule within the muscle [15,16]. In this context two recent literature reviews summarized the effects of compression on metabolite clearance to be controversial [17,18]. In this regard, especially clothing with a high level of compression (>30 mmHg) has not been investigated regarding its effects on blood flow and glucose uptake during recovery.

FIGURE 1.13.2 An extract from an article in a scientific journal of a research report written by scientists. It is strictly structured with date of publication, pertinent title, names of authors and their credentials, and starts with an abstract (summary). The report that follows is not all shown here. It uses the convention of: introduction, method, results, analysis, conclusion and ends with a reference list.

You may find it difficult to access scientific journals because many journals require subscriptions or financial membership. However, some journals are free. Figure 1.13.3 below provides ideas for accessing scientific journals.

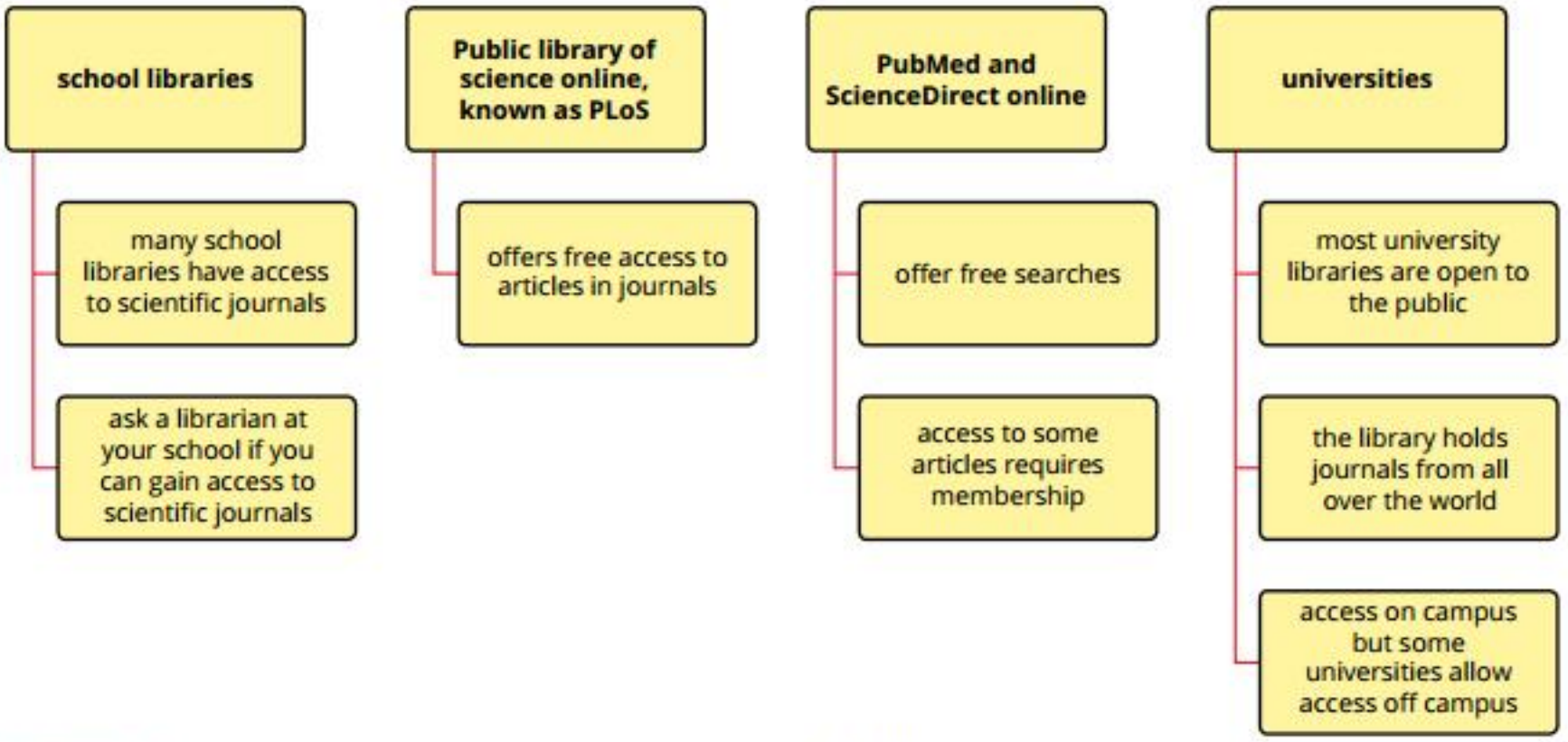

school libraries
many school libraries have access to scientific journals
ask a librarian at your school if you can gain access to scientific journals
Public library of science online, known as PLoS
offers free access to articles in journals
PubMed and ScienceDirect online
offer free searches
access to some articles requires membership
universities
most university libraries are open to the public
the library holds journals from all over the world
access on campus but some universities allow access off campus

FIGURE 1.13.3 Suggestions and information on accessing scientific journal articles

Scientific articles are excellent sources of information, but also have their drawbacks as sources for the research investigation. Table 1.13.3 outlines some advantages and disadvantages of sourcing and using articles in scientific journals for the research investigation.

TABLE 1.13.3 Advantages and disadvantages of scientific articles

Advantages	Disadvantages
• primary source • written by experts with credentials listed • authoritative information (peer-reviewed) • current relevant information • logical, organised layout • contains an abstract that summarises all information in the article and saves you reading time (if you do not see the information you want in the abstract, the article is not relevant) • provides introductory comments that review the literature of the field and place the research focus of the article in context	• information is complex and challenging to understand with specialist terms • advanced processing and analysis of data can be difficult to understand • may be time consuming to read and analyse • well established concepts do not have recent published articles • may have a very narrow focus • may be difficult to locate and gain free access to • sometimes (rarely) written in a language other than English

Books and physical publications

Secondary sources such as good science magazines and books are valuable sources of secondary information.

The first source you should use is your textbook. The language and concepts are presented specifically for high school students and have been carefully reviewed for accuracy. In addition, the textbook addresses the syllabus objectives. Non-fiction books and magazines may be commonly-used resources for the research investigation. Common commercial science magazines you might find as print copies in your school library include *The Helix*, *Cosmos*, *Scientific American* and *New Scientist* (Figure 1.13.4). Table 1.13.4 outlines some advantages and disadvantages of non-fiction books as sources for your research.

FIGURE 1.13.4 A science magazine you might find in your school library

TABLE 1.13.4 Advantages and disadvantages of book and magazine resources

Advantages	Disadvantages
• may be written by experts • potentially authoritative information • logical, organised layout • content is relevant to the topic • contain table of contents and index to help find relevant information • easily located in libraries • written in language that is understandable • some are available as digital versions	• secondary source • may have more bias than primary sources • may not have been published recently • usable by only one person at a time • some are only available as a print copy • may be slower and more inaccessible than an online source

Searching online

Online sources include scientific organisations such as CSIRO, NCBI, AIMS, NIH, Bioscience (Oxford Academic), university publications and credible media sources. Many government and privately funded science organisations publish material, along with not-for-profit scientific organisations. Websites may direct you to magazines and scientific journals, such as those described above, the news, podcasts, blogs and videos (institutional, company and personal).

Information located on the internet requires very careful scrutiny. The openness and ease of publishing on the internet means that information may not be valid and reliable. A source such as Wikipedia can be useful as an accessible and free encyclopedia but not as a reliable or a primary source. It may be useful as a starting point and a way to find external credible sources, such as published scientific papers.

Use the earlier guidelines to help you evaluate sources. Table 1.13.5 outlines advantages and disadvantages of locating and using information online.

When searching for relevant information, you need appropriate search terms to enter in a search engine. These tips can help your search.

- Break your search statement into concepts and keywords.
- Find synonyms, other related terms and concepts that apply to the topic.
- Create concepts of 1–3 words to enter in the search engine.
- Try different combinations of terms.
- Do not settle for the first sites on the list or your first attempt—look beyond the first page of results.
- Look through the results for sites from science organisations and research institutions (e.g. CSIRO, WEHI, NIH; .gov, .org), universities (.edu), science search engines, and science journals and magazines.

TABLE 1.13.5 Advantages and disadvantages of online resources

Advantages	Disadvantages
• quick, easy access to find a range of sources • access to hard-to-find information • international access to a multitude of websites • information is potentially more up to-date • may be interactive and use audio-visuals to enhance understanding • choice of text-based material, animations, images and videos to suit different learning styles	• may not be reliable, valid or credible • can easily side-track with non-relevant information • a lot of 'junk' sites and potentially more biased material that may be out of context or edited • need to discern a list of search engine results to find the most useful sites • cannot always tell how up-to-date the information is or where it comes from • often is not the original source • information may not be well ordered • requires wi-fi and a suitable device

Overview of resources

Your textbook should be your first source of reliable information and other information can be checked for consistency with the textbook. Articles published in journals and magazines often present findings of new research, which may or may not be confirmed later, so be careful not to treat such sources of information as

established fact. Scientific journals are peer-reviewed (critically reviewed by other specialist scientists), which gives them more credibility than other sources.

It is important that sources used in a research investigation are credible and easily accessible because you have limited time for the research.

Forming a question

The claim is used to develop the research question investigated in the assessment task. The variables inferred in the claim may not be measurable or directly observable, and therefore they may not be scientific. Your investigation will aim to have a scientific approach.

To help to develop the research question, break the claim into possible scientific elements, each of which could be used to form a question (Table 1.13.6 below). Separating the claim into elements, such as related terms and concepts, enables you to formulate variables, measurable items and questions, and to identify scientific elements of the claim that are contained in the syllabus unit.

Based on these elements, formulate a number of questions related to the claim and record all in your journal. You will need to understand the dependent and independent variables, and the relationship between the variables, to enable a response that will evaluate the validity or the reliability of the claim. The research question must identify the dependent variable and may also specify the independent variable, and one or more controlled variable(s).

Some guidelines to help form a research question include:

- identifying a dependent variable that can be linked to the claim or rephrasing the claim into something measurable.
- developing an element of the claim into the independent variable or identifying it in the stimulus material.
- phrasing a question to ask if the independent variable will influence, cause or correlate with the dependent variable.
- writing a few different questions.

Usually, the questions improve as you write more, which allows the formulation of a more developed research question. At this stage, these questions can still be quite broad. As they are refined, the focus of the research question needs to become very narrow so that the research report can be completed within the assessment conditions.

The process to developing a research question is often cyclical (Figure 1.13.5).

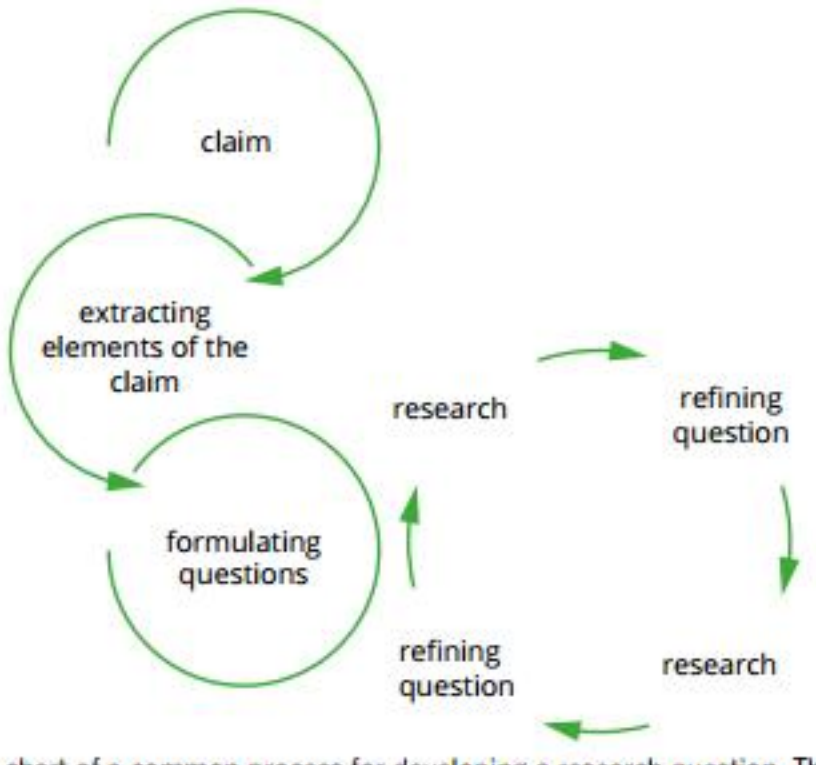


FIGURE 1.13.5 A chart of a common process for developing a research question. This cycle can be repeated as many times as necessary until a scientist is satisfied with the investigation question.

Formulate some broad questions first. See examples of questions formed from an element of each claim when it was refined in Table 1.13.6.

Follow up by refining a formulated question into a research question with variables identified (Table 1.13.7).

TABLE 1.13.6 Three examples of questions formulated from a claim

Example Claim 1: Plastics are bad for the environment.		
Scientific Element	**Elaboration of the claim**	**Formulated questions**
Plastics are made from crude oil.	About 4% of the crude oil is used to make plastic.	Can the efficiency of the distillation process be increased?
	Hydrocarbons are collected by distillation.	
	Non-renewable nature of the resource.	Can reactants in the plastics process be made from something other than crude oil?

Example Claim 2: Natural shampoos are better than commercial shampoos.		
Elements	**Elaboration of the claim**	**Formulated questions**
Natural shampoos are as effective cleaning products as commercial shampoos.	Shampoo effectiveness can be evaluated by comparing the surfactant properties of the shampoos.	Does a greater reduction in the surface tension mean that the shampoo is more or less effective?
	Surfactants have a non-polar aliphatic chain and polar head.	Do natural shampoos work in the same way as synthetic shampoos?
	Properties: • surface tension • wetting ability • solid content • detergency • foaming ability	Is the volume of foam related to effectiveness of cleaning?

Example Claim 3: Green chemistry is clean chemistry.		
Elements	**Elaboration of the claim**	**Formulated questions**
Green chemistry	Atom economy	Can the chemical yield of a reaction be increased?
		Does a higher atom economy mean a greener product?
		Is it possible to make the same product in different ways?

Refining the research question

One of the questions in your journal will become your research question. As you conduct research into the independent and dependent variables, new information will refine the question, so it is more specific.

Since the Forming and Finding criterion assesses the development of the research question from the claim, it is important to record your development of conceptual understanding and the knowledge you gain about the relationships between variables.

Table 1.13.7 compares a formulated question with a developed research question. Always get the teacher to assess your research question.

TABLE 1.13.7 Development of the original questions formulated into research questions

Example Claim 1: Plastics are bad for the environment.		
Formulated question	**Refined research question**	**Variables**
Can the efficiency of the distillation process be increased?	If the number of stages within the fractional distillation tower is increased to collect fractions with a smaller temperature range, could polymers be made more environmentally safe?	*Dependent:* Environmental safety of the polymer. *Independent:* Number of stages in the distillation tower **OR** Fraction size
Can reactants in the plastic process be made from something other than crude oil?	Can the monomers that are used to generate the plastics used in polyurethanes be generated from a renewable resource?	*Dependent:* Number of monomers *Independent:* Renewable resource

Example Claim 2: Natural shampoos are better than commercial shampoos		
Formulated question	**Refined research question**	**Variables**
Does a greater reduction in the surface tension mean that the shampoo is more effective?	Are natural shampoos better cleaners than synthetic shampoos based on the surface tension, wetting ability, solid content, detergency and foaming ability?	*Dependent:* Cleaning efficiency *Independent:* Shampoo type (natural/ synthetic)
Do natural shampoos work in the same way as synthetic shampoos?	Do the chemicals in natural shampoos create micelles that have a non-polar aliphatic chain with a polar head that increase the solubility of the oily substances in water?	*Dependent:* Solubility of (choose a substance) *Independent:* Shampoo type (natural/ synthetic)

Example Claim 3: Green chemistry is clean chemistry		
Formulated question	**Refined research question**	**Variables**
Can the chemical yield of a reaction be increased?	Would increasing the temperature in the Haber process (used to manufacture ammonia) to shift the equilibrium towards the products increase the overall greenness of the ammonia production process?	*Dependent:* Greenness of the Haber process *Independent:* Temperature
Does a higher atom economy mean a greener product?	Can improving the chemical yield of the synthetic reactions through higher atom economy create a greener manufacturing process?	*Dependent:* 'Greener' manufacturing process OR Chemical yield *Independent:* Atom economy
Is it possible to make the same product in different ways?	Can the polymers be made from non-petroleum sources and so increase the environmental safety level of plastics?	*Dependent:* Environmental safety of plastic using the resin identification code. *Independent:* Source of polymer (resin identification code).

Your research investigation report requires a considered rationale for the research question, displaying its clear development from the claim. Outline this step-by-step development, justifying the steps using scientific concepts, knowledge and understanding.

Therefore, a research question must explicitly identify the dependent and independent variables. The research question must be connected to the considered rationale and the topic of study.

Putting it all together

The structure of the research report is not mandated by the assessment specifications but needs to be presented using appropriate report conventions, such as appropriate headings and language and acknowledge the source of the information using referencing conventions (refer to Module 1.8).

Possible structures include headings such as:

- Claim, Rationale, Research question, Introduction or Background, Analysis and Interpretation, Conclusion and Evaluation.
- Or the headings of the instrument-specific marking guide (ISMG) criteria as stated in the QCAA syllabus: Forming and Finding, Analysing, Interpreting and Evaluating.
- Or you may elect to organise your report in a different but logical order.

The report should use scientific writing style, following the advice and directions presented in Part A Module 1.8 Scientific writing.

The sample research investigation report that follows covers the ISMG criteria for Forming and Finding, and may be used as a reference tool. Use this as a model for reference as you write your own report.

Claim:

We are running out of effective antibiotics.

Rationale:

The effectiveness of antibiotic treatments is decreasing worldwide, reducing the ability to eradicate and cure infectious diseases. The claim, 'We are running out of effective antibiotic treatments'. has important aspects to investigate. Antibiotics are chemicals that prevent bacterial cells from multiplying, enabling the human immune system to fight infections. Penicillins are the original group of antibiotics, first developed by Dr. Alexander Fleming (Gora, 2023).

The bacterial diseases of pneumonia, meningococcal and gonorrhoea use penicillin-based antibiotics as treatments. They were chosen for accurate comparisons of antibiotic effectiveness. Pneumonia is treated with Amoxicillin, which increased its ineffectiveness by 30% of patients in a study conducted by Centres of Disease Control and Prevention. Meningococcal typically uses the Cephalosporins antibiotics, Gonorrhoea treatment also includes Cephalosporins, and resistance has progressively developed to the antibiotics (Shaskoskiy, 2019). It is projected that 10 million people with meningococcal will die each year across the world (WHO, 2020) due to this resistance.

The number of effective antibiotics is the dependent variable of this claim, in relation to the independent variable of how many years antibiotics have been used to treat bacterial diseases.

The aspect of 'running out' refers to the growing ineffectiveness of antibiotics (Willacy, 2020). This claim is supported by World Health Organization (WHO) reports in 2017, 2020 and 2023, detailing the growing threat of antibiotic resistance. The reports identified the increase in short-term solutions that are not sustainable for the future, and found very few long-term development plans for new antibiotics.

The 'We' of the claim is the human race. The overuse of prescribed antibiotics is predicted to negatively impact humans as pathogens build an immunity to existing antibiotics. Doctors globally identify antibiotic over-prescription as accelerating the natural immunity evolved over time by bacteria.

Antibiotic or antimicrobial resistance (AMR) occurs when bacteria develop the ability to resist antibiotics, making bacterial diseases untreatable. Antibiotic resistance is responsible for at least 1.27 million fatalities worldwide. Contributing causes include the over-prescription of antibiotics, overuse in livestock and agriculture, poor infection control, poor hygiene and sanitation, and absence of new antibiotics development (WHO, 2023).

Research question:

How has the overuse of penicillin-based antibiotics (Amoxicillin and Cephalosporins) impacted the treatment of pneumonia, meningococcal and gonorrhoea when analysing the patient's ability to eradicate these diseases?

1.14 Analysing

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- identify the variables in the research question
- determine the evidence required to evaluate the validity of the research question
- identify relevant patterns and trends in the data
- explain the patterns between the two variables
- identify the limitations or gaps that exist in evidence.

ANALYSING EVIDENCE

The components of the Analysing criterion are outlined in Module 1.14, Table 1.14.1.

The ISMG for the Analysing criterion is assessed in relation to the identification of sufficient and relevant data that enables the research question to be answered.

Analysing evidence that is sufficient and relevant involves a description of relationships between dependent and independent variables. The data should be presented from more than one source and the overall trend from two or more authors needs to be identified.

The ISMG includes the identification of trends, patterns and relationships in the data and the limitations of the evidence. A trend or pattern is the general change in one characteristic with respect to another. In science, these are often (but not always), represented in the form of a graph with the dependent variable on the y-axis and the independent variable on the x-axis. In Biology, different types of graphs are used depending on the nature of the data. The graphing information in Module 1.6 of Part A provides examples of all these.

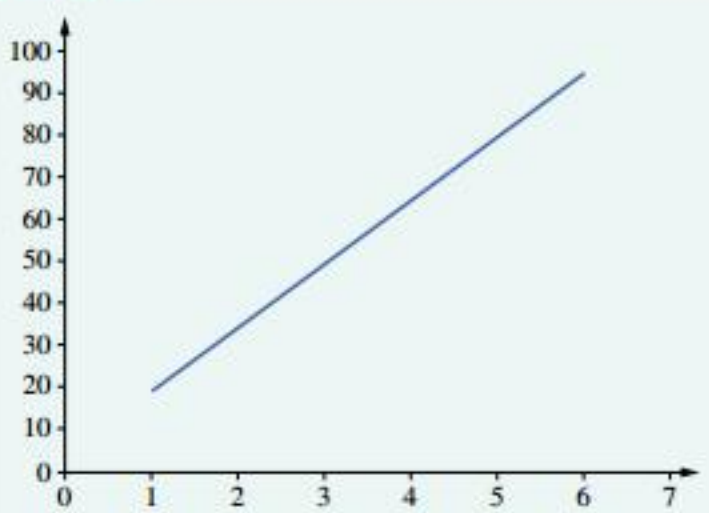
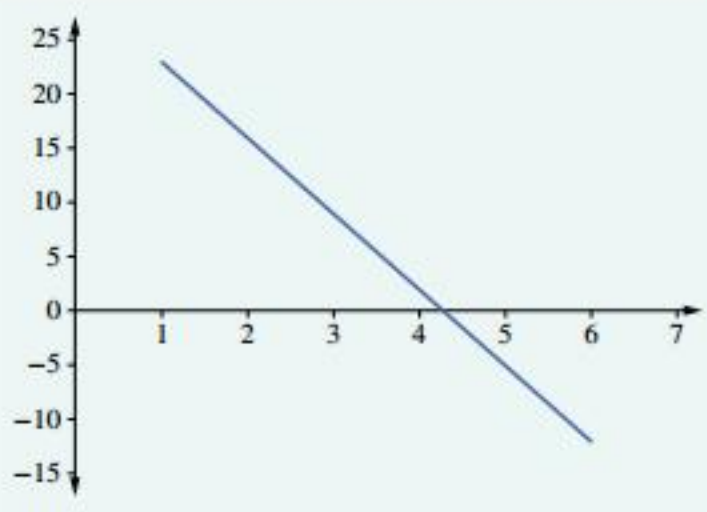
Most commonly, especially when identifying relationships between variables, a scatterplot of data points is plotted and then a trend line (line of best fit) drawn through the points. Examples of the resulting line graphs are shown in Table 1.14.1, along with the term used to describe each graph category.

The first thing to notice about the trend is the shape of the graph. If there is a relationship between two variables the trends can typically be put into one of five categories: linear, power, inverse, exponential and logarithmic.

In addition to the type of relationship, there are other factors to describe. For instance, does the dependent variable increase as the independent variable increases? Does the increase get bigger, as is the case with an exponential growth or does it plateau, consistent with a logarithmic change?

Examples of the different shapes are shown in Table 1.14.1.

TABLE 1.14.1 Examples of different graphical relationships: linear and non-linear (power, inverse, exponential, logarithmic)

Type of graph	Examples
Linear	 

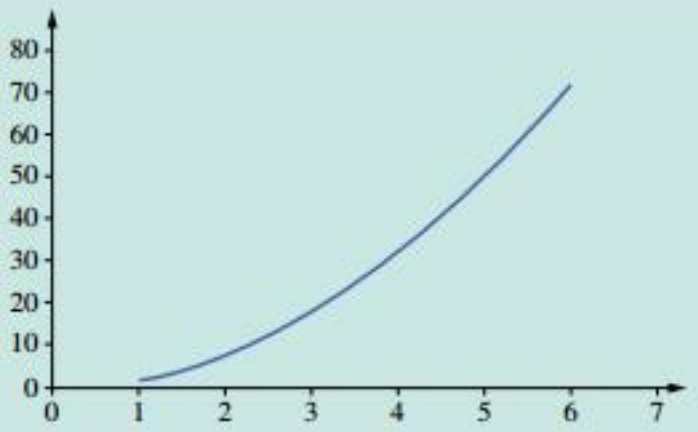
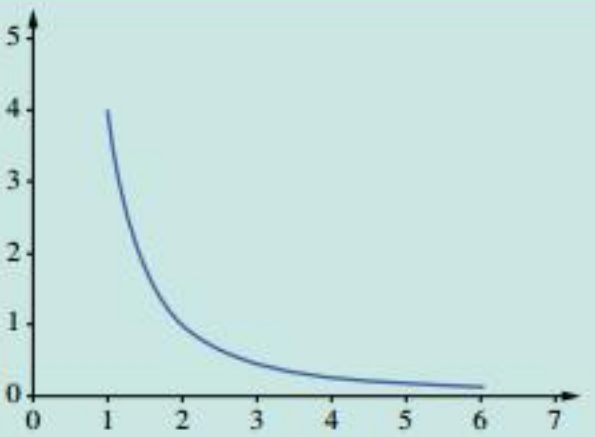
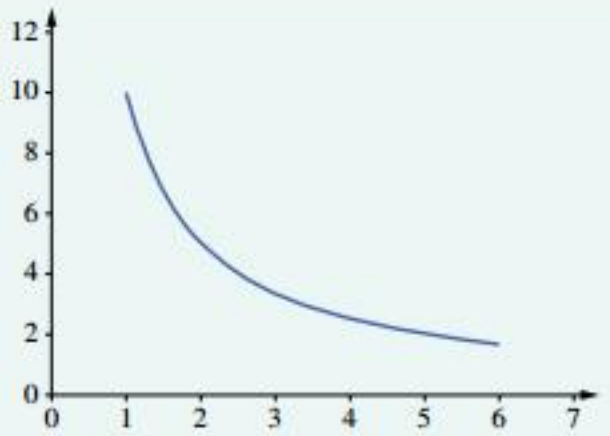
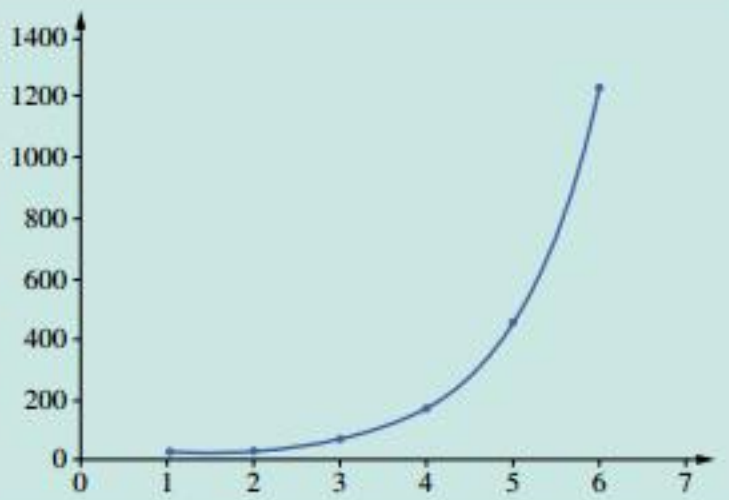
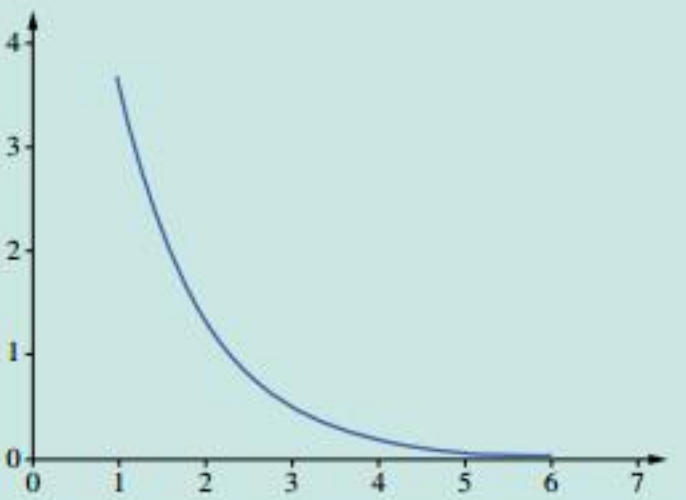
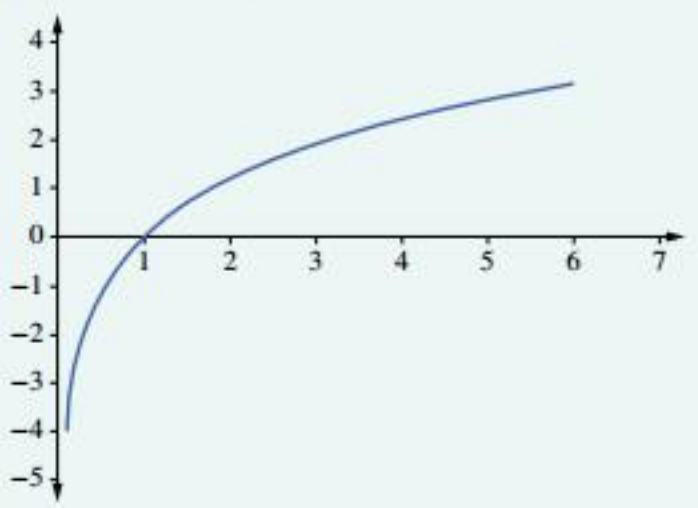
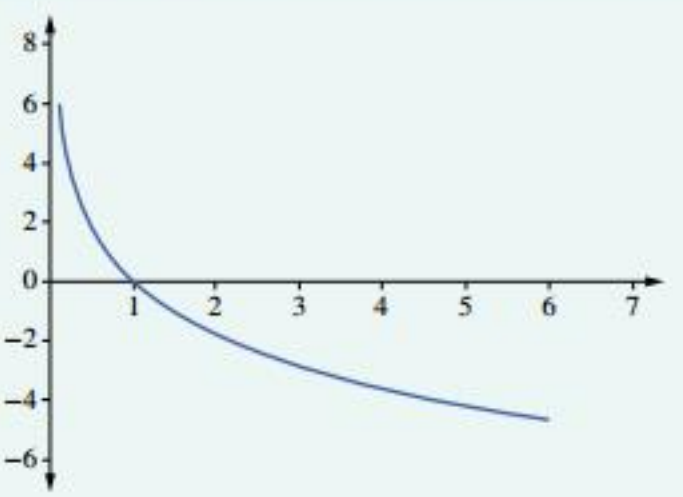
Type of graph	Examples	
Power		
Inverse		
Exponential		
Logarithmic		

Figure 1.14.1 shows a comparison of two linear trends. The graph shows the change in the mass of products as a function of time. While a trendline for each is shown no error bars are included for this discussion. When analysing relationships, it can be necessary to compare the effects of changing conditions on the dependent variable. As well as determining the gradients of the curves, key features such the *x*- and *y*-intercepts are important. While both linear graphs show that as time increases the mass of product increases, the rate of increase or gradient is greater for the dashed line. The dashed line passes through the origin (point 0, 0), while the solid line extrapolates at a negative intercept.

Note, the Analysing criterion identifies the relationship but does not assess the reasons for the relationship. That will be assessed in the Interpreting criterion.

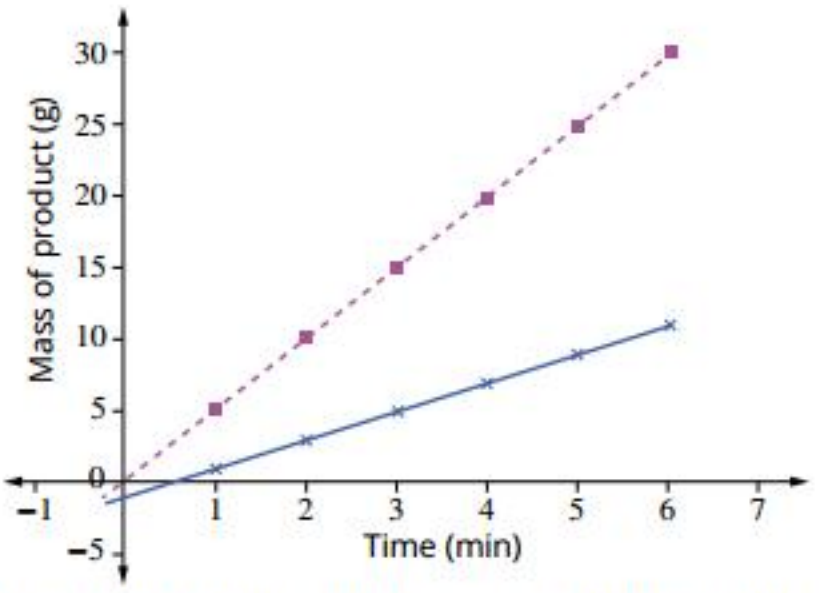


FIGURE 1.14.1 Comparison of the formation of product of a reaction under different conditions. In biology, it could represent enzyme reactions under different conditions, such as two different temperatures, although a negative would not be expected unless it was a measurement error.

Although a graph is the common form to identify relationships between variables, it is not the only scientific convention to use. A dataset can also be described by calculating the percentage difference. For example, when comparing the degree of biodegradability of two plastics over a 28-day period, this method can help quantify the relative differences. It is also important to consider that data may be presented from more than one source. The overall trend should be identified by comparing and synthesising findings from several authors to ensure a comprehensive understanding of the relationship.

LIMITATIONS OF THE DATA

All studies are affected by limitations. When analysing the data, it is important to identify the limitations. Limitations can often be grouped into three categories:

- elements of the methodology
- study design that impacts the interpretation of the research results
- differences between the studies or sources used to investigate the relationship.

Examples of common methodological issues include:

- Methods, instruments and techniques used to collect the data—While it is unlikely that the students will have experience in using many of the instruments used to collect the scientific research data, any perceived or identified limitations in the methodology should be outlined in the discussion section of the research paper. This could include, but is not limited to, the instrument's level of detection.
- Selection of samples—This can result in a selection bias if the sampling method results in the selection of samples that do not reflect the general population or population concerned.
- Insufficient sample size for statistical measurements—It is critical to have a sufficient sample size to draw valid conclusions. More precise results can be obtained if a larger number of samples are collected, which also enable the results to be generalised to a larger population. In contrast, if the sample size is too small it can be difficult to identify significant relationships in the data.
- Lack of previous research studies on the topic—If the research cited is from a limited number into the issue, the lack of available or reliable data can make it difficult for firm conclusions or to make connections between the studies, or the findings and the real world. However, such studies should not necessarily be dismissed. Science is always evolving and developing new areas of research. It should be recognised that some research will be at the forefront of a new field without a body of previous research, or from a country with limited means for scientific research, for example, the study of antibiotic resistance in Bhutan

described in the sample research report. As such, it offers a good opportunity to discuss limitations and improvements.

- The differences between the studies or sources used to investigate the relationship—While different studies may have the same dependent and independent variables, they are unlikely to have used the same methods or materials. Additionally, even if studies elucidate the same relationship between variables, they may show differences in the degree or extent of the relationship. For example, an enzyme that is shown to promote a reaction, increases the yield but one source used a different reaction condition or temperature to a second source. Note that you will be using data from 3–5 different sources to determine the overall trend, and it is unlikely that these researchers used exactly the same methods or materials.

When reading the research papers, refer to the collection of sufficient and relevant raw data in Part B Module 1.10. This will help evaluate the analysis of the data and identify limitations of the methodology.

As with identification of the trends and patterns, the Analysing criterion only assesses the identification of the limitations of the research. The role these limitations played on the data collection and potential impact of the study, including the inability to generalise the research findings will form part of the Evaluating criterion

The report should also suggest possible ways to overcome the limitations in future studies, such as alternative methodologies and ways to avoid issues.

Ways to discuss the limitations

When identifying limitations of the study there are ways to introduce the discussion on these limitations. Remember to be concise and stay within your word limit of 2000.

- Possible limitations of the study include …
- The first is the…The second limitation concerns the…
- The empirical results reported by … should be considered in the light of some limitations.
- This research is subject to several limitations.
- The primary limitation to the application of these results is…
- The design of the current study is subject to limitations.
- The major limitations in this study that could be addressed in future research are first: the study focused on …. second: the study …
- The same overall trend was identified from three sources. However, a limitation is that their methodologies were not identical, in that …

SUFFICIENT AND RELEVANT

When deciding whether the sources are relevant or sufficient, refer to the start of this module. The Analysing criterion uses the terms 'sufficient' and 'relevant'. For this section, the terms refer specifically to assessment of the evidence. Was the data that was analysed 'enough and connected with the investigation' to enable the research question to be fully answered?

Analysing: putting it all together

When writing the research report, it is important to evaluate the report against each criterion in the ISMG to assess the quality of the response. The Analysing criterion is focused on the evidence that is collected, identified and described patterns or trends that are evident, and connected to the research question. Therefore, it is critical that a student identifies all aspects of the research question that need to be answered. This will enable effective searching for and selection of information related to these aspects.

Once the evidence has been found, the trends need to be identified and used to answer the research question. Remember, that the Analysing criterion is only assessed against identification of the patterns and identifying the limitations of the research. So, to get the highest mark, the report needs to collect and analyse enough information to answer the research question.

The sample research investigation report that follows shows the analysis of evidence for the research question developed from the claim and covers the ISMG criteria. It may be used as a reference tool. Use this as a model for reference as you write your own report.

Analysing

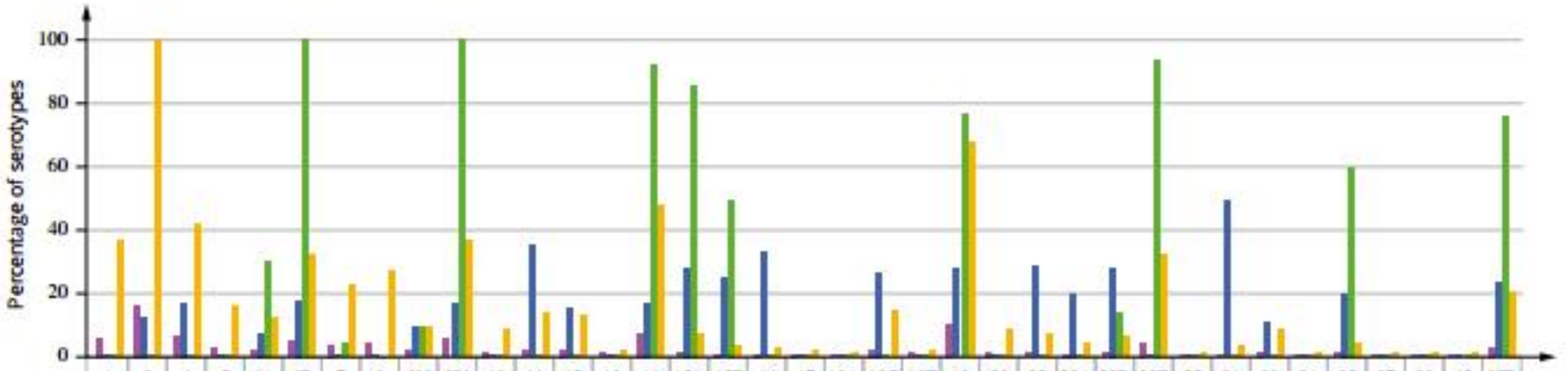
Secondary research was conducted to find medical studies that related the use of penicillin-type antibiotics to development of resistance (dependent variable) over a period of time (independent variable) when used for treatment of three specific bacterial diseases.

Evidence 1 – Pneumonia

The rate of penicillin resistance amongst clinical isolates of *Streptococcus pneumoniae* increased by 35.7% in Spain. The Alexander Project conducted a multicentre study from 1992 to 2002 to examine the evolution of antibacterial resistance in community respiratory bacteria (Felmingham, 2005). In a later study, the increased levels of antibiotic resistance were investigated for 638 cases of community-acquired pneumonia (CAP) resulting from *Streptococcus pneumoniae* (Aspa, 2004). It was identified that the overall incidence of multiple-antibiotic resistance within *S. Pneumoniae* isolates is increasing. As evident in Figure 1, the graphs display the *S. Pneumoniae* serotypes in relation to relative frequency, resistance and associated mortality rate.

The data identifies that since the Alexander Project, there is an increase in penicillin resistance, along with an increased associated mortality rate (EXITUS). Fifty of 409 (12.2%) patients with CAP caused by penicillin-susceptible pneumococci died, along with 12 of 65 (18.5%) patients infected with strains that were resistant to penicillin.

Other health factors were considered and found to also influence mortality. Limitations analysed during the data collection included a gender bias with more males, an age bias with the mean age being 61.58 years, plus old and outdated methodologies. This limited the data because the elderly have higher disease susceptibility compared to younger participants. The study was conducted from January 1999 to April 2000, so it does not represent the data today.



	1	3	4	5	6A	6B	7	8	9N	9V	10	11	12	13	14	15A	15F	16	17	18A	18C	18F	19	20	22	23A	23B	23F	25	31	33	34	35	37	38	42	NT
RELAT FRE	5.8	16.9	6.6	2.5	2	5.2	3.6	4.2	1.6	5.8	1.4	2.2	2	0.3	7.5	1.1	0.6	0.5	0.3	0.2	2.4	0.3	10.7	1.4	1.1	0.8	1.1	5	0.2	0.6	1.4	0.2	0.8	0.2	0.2	0.2	3.3
EXITUS	0	13	16.7	0	7.7	18.2	4.3	0	10	16.2	0	35.7	15.4	0	16.7	28.6	25	33.3	0	0	26.7	0	27.9	0	28.6	20	28.6	0	0	50	11.1	0	20	0	0	0	23.8
PEN RES	0	0	0	0	30.8	100	0	0	10	100	0	0	0	0	91.7	85.7	50	0	0	0	0	0	76.5	0	0	0	14.3	93.8	0	0	0	0	60	0	0	0	76.2
n	37	108	42	16	13	33	23	27	10	37	9	14	13	2	48	7	4	3	2	1	15	2	68	9	7	5	7	32	1	4	9	1	5	1	1	1	21

FIGURE 1 Distribution of Streptococcus *pneumoniae* serotypes, according to relative frequency, resistance to penicillin, and associated mortality rate (EXITUS). Data are percentage of serotypes, unless otherwise indicated. Where *EXITUS* is overall associated mortality rate; *n*, number of patients for each serotype; *NT*, nontypeable serotype; *PENI RES*, resistance to penicillin (MIC, ≥0.12 μg/mL); *RELAT FRE*, relative frequency of each serotype. (Aspa, 2004)

A more recent study in India identified *S. pneumoniae* to rank as the fourth-most lethal pathogen globally in terms of fatalities associated with or attributable to resistance (Kulkarni, 2023). This study conducted clinical trials on specimens collected from 2018 to 2021 and tested with penicillin and other antibiotics. The standard laboratory protocol used, known as AST (antibacterial susceptibility testing), determines if the pathogen is susceptible or resistant to an antibiotic.

As can be seen in Figure 2, penicillin susceptibility ranged from 0% in 2018 to a peak of 37.5% in 2019, and then to 30% in 2021. This suggests that over four years the resistance for *S. pneumoniae* bacteria to penicillin had increased from 0% to between 30% and 37.5%. Between 2019 to 2021, the study was impacted by SARS-CoV-2 infections and precautions. This limitation could explain the small drop in penicillin resistance for 2000 and 2021. Another limitation is that the research applies only to one country over four years and had a small sample size of 86 isolates.

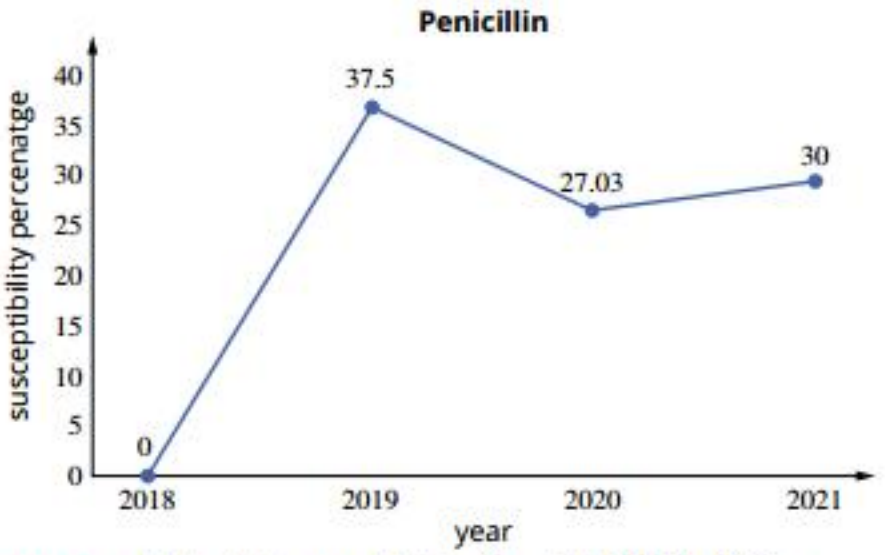
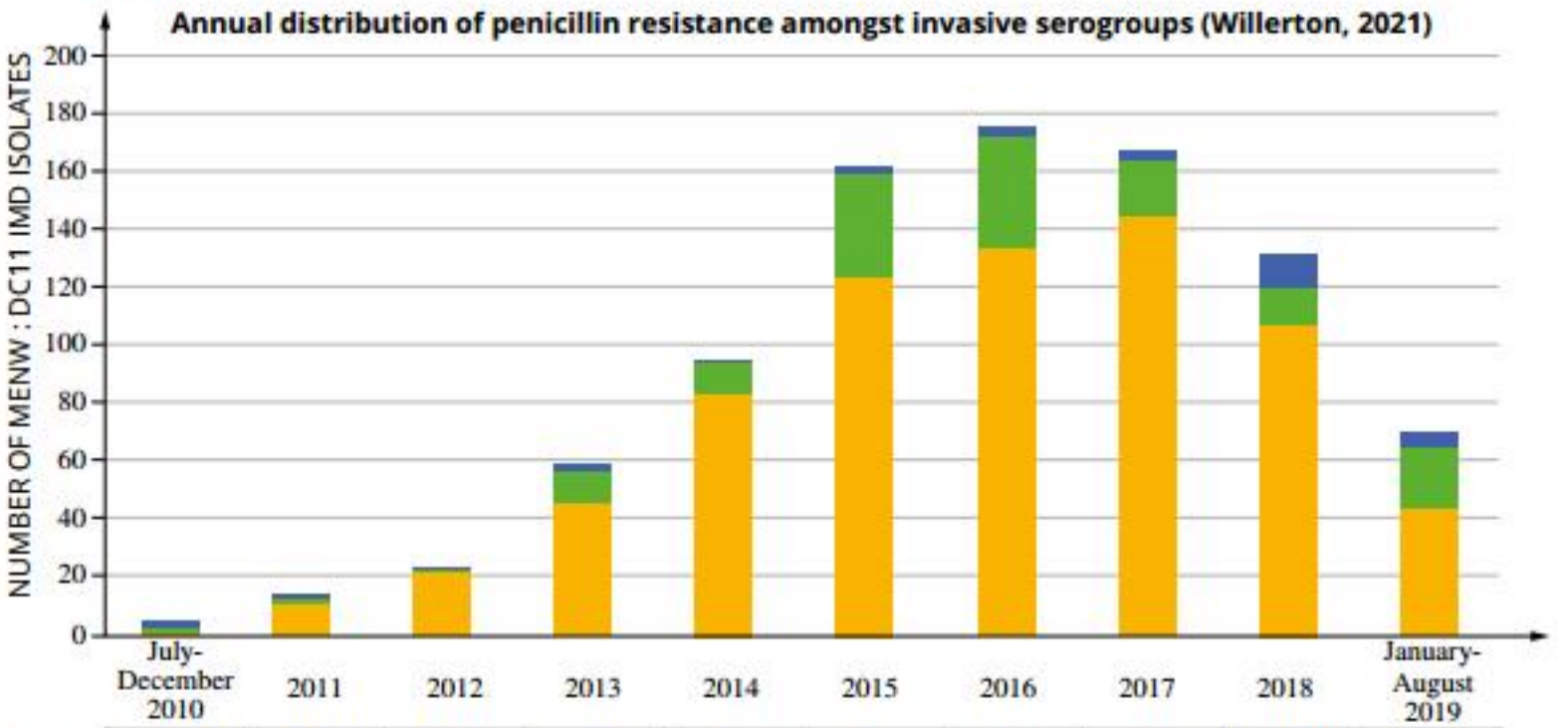


FIGURE 2 Antibacterial susceptibility of penicillin 2018 to 2021 (Kulkarni, 2023)

Evidence 2 – Meningococcal



	July-December 2010	2011	2012	2013	2014	2015	2016	2017	2018	January-August 2019
PenR (MICs > 0.25mg/L)	1 (25.00)	0	1 (4.35)	1 (1.72)	0	1 (0.62)	2 (1.14)	2 (1.20)	12 (9.09)	5 (7.14)
PenI (MICs 0.094–0.125 mg/L)	1 (25.00)	2 (15.38)	1 (4.35)	11 (18.97)	11 (11.70)	37 (22.98)	39 (22.29)	20 (11.98)	13 (9.85)	21 (30.00)
PenS (MICs ≤ 0.06 mg/L)	2 (50.00)	11 (84.62)	21 (91.30)	46 (79.31)	83 (88.30)	123 (76.40)	134 (76.57)	145 (86.83)	107 (81.06)	44 (62.86)

FIGURE 3 Annual distribution of penicillin resistance amongst invasive serogroups (Willerton, 2021)

While only 2.8% of isolates display resistance, resistance has increased over the years by 35.88%. Conducted by the Meningococcal Reference Unit, the study has analysed the increase in penicillin-resistant Invasive Meningococcal Disease (IMD) (Willerton, 2021). Through examining IMD cases in England from July 2010 to August 2019, the data was determined from patients undergoing genome sequencing and antibiotic susceptibility.

Figure 3 exhibits the number of Penicillin-Susceptible isolates: Standard Exposure (PenS), Penicillin-Susceptible: Increased Exposure (PenI) and Penicillin Resistant (PenR). The results revealed that out of the 897 MenW:cc11 isolates, 716 (79.8%) were penicillin-susceptible with standard dosing regimen, 156 (17.4%) were penicillin-susceptible, increased exposure, and 25 (2.8%) were penicillin-resistant. The graph also indicates the increase of PenR patients, identified among 6 sublineages and a singleton, from 2010 to 2019. In 2018, there were 12 PenR isolates which was the highest annual proportion of isolates within the 8½ years of examination. This is in comparison to 2010's isolates where only 1 was PenR. Laura Willerton of PLOS ONE stated that IMD antibiotic resistance is a result

of altered PenA alleles and additional mutations along with over-prescription (HealthDirect, 2021). A limitation of the data collection was that it was collected only in England.

The results represent a population sample living with similar conditions and health care in comparison to other cities around the world.

Evidence 3 – Gonorrhoea

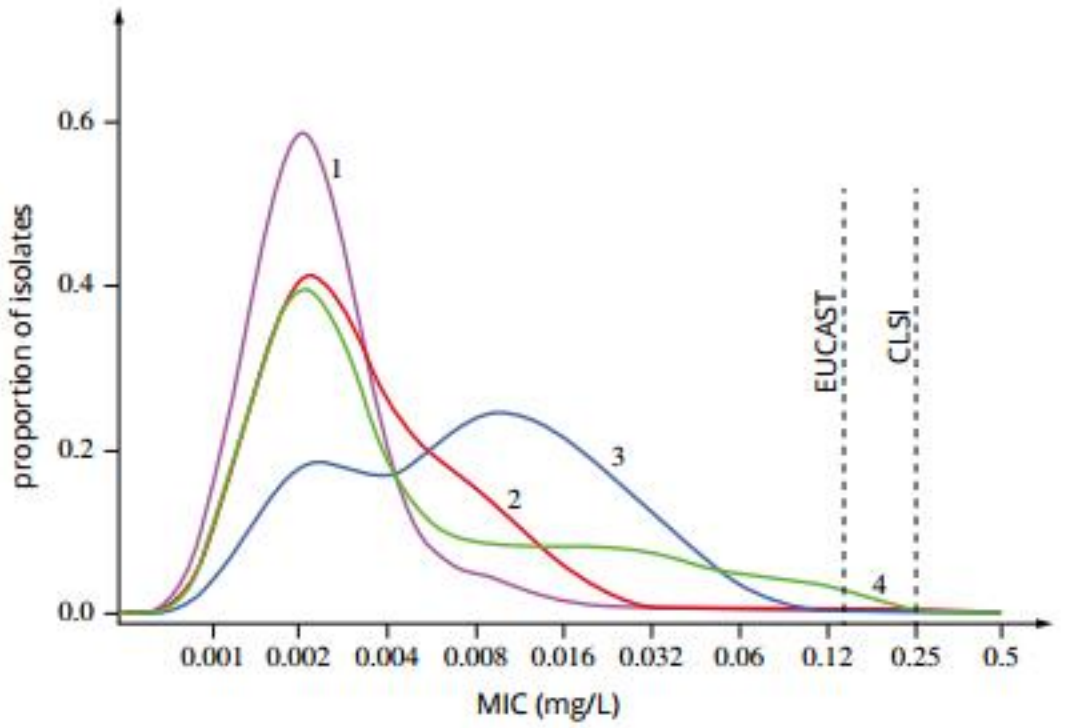


FIGURE 4 Distributions of *N. gonorrhoeae* isolates with different genetic determinant profiles. Wild-type isolates (1), isolates with mutations (2), isolates with mutations in three or four chromosomal genes (3), and isolates carrying blaTEM plasmids (4) (Shaskoskiy 2019)

The resistance determinate was exhibited by 72.4% of isolates. The study by Boris Shaskolskiy (2019) aimed to investigate the phenotypic susceptible and resistance determinates of gonorrhoea isolates to benzylpenicillin. In Russia, 522 participants were analysed incorporating the agar dilution method. Figure 4 displayed that resistance to benzylpenicillin was at 7.7% in isolates, with 72.4% of isolates exhibiting resistance determinate with the Asp345 insertion. A distinctive feature of *Neisseria gonorrhoeae* is its ability to rapidly accumulate different mutations to acquire resistance against the antibiotics used for its treatment. The molecular determinants associated with the resistance of *N. gonorrhoeae* to penicillins involve both chromosomal mutations and the presence of the bla_{TEM} plasmid encoding beta-lactamases (penicillinases). The human use of antibiotics has increased the number of genetic mutations and plasmids that carry mutations in the gonorrhoea bacteria, leading to possible faster development of resistance.

The Russian study is supported by Tshokey (2015), with a study of antibiotic resistance in *N. gonorrhoea* in Bhutan. They concluded that resistance against ciprofloxacin, penicillin, tetracycline and nalidixic acid were 85.1%, 99.2%, 84.8% and 99.7% respectively of 524 patients from two hospitals due to over-prescription.

While sample sizes at 522 and 524 were adequate, there were limitations present during the data collection and testing for both studies. In the Russian study, samples were anonymous for ethical reasons, so some potential for data analysis was lost, such as age, gender and complicating medical conditions. A limitation noted for the Russian assay was that only a restricted number of genetic markers relevant to antibiotic resistance could be tested. The Bhutan study collected more personal data but noted the lack of uniform medical protocols in that country and that 58% of their patients were lost to follow up once they left hospital. Also, that more reliable clinical susceptibility testing methods were required in future studies.

1.15 Interpreting

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- evaluate the research process
- understand the steps and requirements for a justified argument
- identify the steps necessary to develop a justifiable conclusion.

The ISMG for the Interpreting criterion, is shown in Module 1.14, Table 1.14.1. To achieve the highest mark (5) in the Interpreting criterion you need to address three aspects: create justified scientific arguments, a justified conclusion to answer the research question, and present the results in the form of a scientific report using fluent scientific language and representations.

JUSTIFYING AN ARGUMENT

When constructing a scientific argument, the claim, evidence and scientific reasoning need to be considered:

- The claim is what is known.
- The evidence is how this is known.
- The scientific reasoning is why the evidence supports or refutes the claim.

The QCAA glossary of cognitive verbs defines **justify** as 'to give reasons or evidence to support an answer, response or conclusion; show or prove how an argument, statement or conclusion is right or reasonable'

A scientific argument is a statement supported by multiple pieces of evidence that is measured or observed. The argument links the evidence and reasoning together with scientific facts and knowledge. It is important to recognise that the argument could either corroborate the claim or provide evidence that refutes the claim.

Therefore, the investigation needs to specifically use the information and evidence collected from the research to answer the research question, rather than simply opinions or beliefs that cannot be empirically verified.

The Forming and Finding section outlined terms and concepts related to the topic, which were used to develop the research question. This helped identify variables linked to the research question to be analysed.

The Analysing section of the report identified patterns and trends in the data related to the research topic.

The Interpreting section of the report needs to explain the trends between the variables in order to answer the research question.

Common things to avoid

When writing the justification, it is important to be concise and to the point. Select data and explain how the evidence is linked to the research question. Ignore evidence that has no relevance to the research question.

Be selective about the information discussed because the specifications of the assessment task state that the length of the report is limited to 2000 words or an 11 minutes presentation. The justification needs to emphasise the significance of the research to the research question or topic. How do the trends identified in the analysis section support the conclusion that was drawn. If the data does not directly address the question it should be removed from the report. It is not necessary to use the whole of the data from the research, only use parts of the data that address the research question.

Drawing a conclusion

When writing the conclusion make sure that it explicitly answers the research question. The easiest way to do this is to state the research question and summarise, in up to 150 words, how the evidence supports the research question, using the identified variables. In the summary, refer to the key evidence and authors or figures. In addition, concisely outline key limitations and how they impact the conclusion with respect to the research question.

PUTTING IT ALL TOGETHER

There is no single way of creating a justified argument to answer the research question or write the conclusion. The interpretation of the research collected simply needs to create justified arguments that use the evidence to explain and answer the research question to form a justified conclusion

The sample research investigation report that follows, covers the ISMG criteria and may be used as a reference tool. Use this as a model for reference. The sample shows one way to structure justified arguments that link the research question and evidence, enabling a conclusion to be drawn that is supported by reasoning and scientific understanding. Remember that in your report the conclusion can be placed either before or after the Evaluating section (Module 1.18). For this sample you will find it after, at the end of Module 1.18.

Summary Of evidence

The research task aimed to identify how the overuse of penicillin-based antibiotics impacted the treatment of pneumonia, meningococcal and gonorrhoea bacterial diseases. It was evident that in:

Evidence 1 the overall incidence of penicillin resistance within *S. pneumoniae* isolates is increasing with 35.7% of isolates exhibiting resistance.

Evidence 2 there was an increase annually in meningococcal isolates exhibiting resistance from 1 patient in 2010 to 12 patients in 2018.

Evidence 3 it was discovered that 72.4% of gonorrhoea isolates exhibited the resistance determinate.

The patterns recognised include the growing rate of resistance and the increased rate of resistance mutations (Evidence 2). All three pieces of evidence explored the growing rate of resistance compared to past years. For example, with Evidence 1 in India, resistance went from zero to over 30% in just 4 years and with Evidence 2, the number of isolates exhibiting resistance went from 1 isolate to 12 within 8 years. Evidence 3 also explored the impact of mutations on resistance, with the source saying it contributed to the increasing rate of resistance.

Relationships identified that contribute to resistance include the over-prescription of antibiotics and how the participants' age increases the rate of resistance. The relationship between the dependent variable of resistance and the overuse of antibiotics is observed through bacterial pathogens building immunity against antibiotics over time, rendering them restricted in curing the disease. The other relationship was between the two variables of sample age (IV) and resistance (DV). While older patients have more immune system experience combating the diseases using antibiotics, this prolonged use has resulted in disease pathogens being more resistant in the elderly than younger generations. This was seen in Evidence 1, where the average mean age of 61.58 years had a high resistance rate of 35.7% of isolates.

1.16 Evaluating

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- evaluate the research process
- identify the steps necessary to develop a justifiable conclusion
- evaluate the quality of the evidence
- extrapolate the conclusion to the original claim.

The ISMG for the evaluating criterion, is shown in Module 1.14, Table 1.14.1. It is assessed in relation to evaluating the quality of the evidence, generalising the information to the original claim and suggesting improvements and extensions that relate to the claim.

To achieve the highest mark (5) in the Evaluating criterion you need to evaluate the quality of information used to support the scientific arguments. This requires an explanation of the limitations of the data including an assessment of the statistical quality of the data, such as R^2 value or error bars. In addition, while the focus of the report is to justify a very narrow research question, this section needs to extrapolate the findings to the much wider claim, i.e. if the findings were more widely applicable how this would impact the claim. Finally, what improvements and extensions would strengthen the findings and further justify the answer to the research question.

EVALUATE THE QUALITY OF EVIDENCE

The Analysing section of the report identified limitations in the data. Now, the Evaluating section of the report needs to explain how the limitations identified in the data and the sources used to support the argument, affected the ability to evaluate and answer the research question.

Questions that could be useful to answer when evaluating the reliability of the data include:

- Were the statistical measures of the data good and appropriate?
  This could be through a R^2 value close to 1 or very small error bars in the data trend.
- Was the sample size sufficient to say that the findings were valid?

As the sample size increases, the standard deviation of the results decreases, meaning that the chance of results being skewed by a single outlier is less.

- Were the findings reproducible?
- Were the results of the research investigation corroborated by other studies and research institutions?
- Were the experiments in the study repeated with different techniques and the findings were consistent or independent of the methodology chosen, without a justifiable reason why they might vary?
- Did the result occur only under very specific conditions and were these conditions applicable to the real-world scenario?
- Was the methodology and instrumentation applied appropriately to the situation studied?
- If the study was to investigate numbers significantly greater than the sample, can the findings of the research be applied to the real world?
- Are the theories that the authors used to support their findings still valid?

Do the results fit the accepted theories or equations? For instance, in Figure 1.16.2 one of the trendlines went through the origin. This means that at the start of the experiment the mass of products formed was 0 g; however, the other trendline went through −1 g. This makes no sense in relation to the experimental data and so indicates that the results of this study may not be reliable. In a peer-reviewed journal it is likely that these issues would be identified and discussed by the authors.

It is important not to just repeat the statements earlier in the Analysis but to describe the effect on the investigation.

In addition to evaluating the data analysed, the investigation needs to evaluate if the sources that were used are reliable. An accurate reference list must be appended at the end of the report, using a recognised citation method. Refer to Module 1.8 for writing a reference list.

Evaluating sources for validity and reliability

Determining the validity and reliability of a source can be a challenging task. For some sources, it is easy to find details about the author, evidence and concurrency, while others only contain content and do not offer any other details. Another difficulty is when learning about a new topic or concept, we are all novice learners so it can be challenging to interpret the language and to tell if a source is valid or not.

Examples of questions to consider when evaluating the sources include:

- Who or what organisation commissioned the report?
- What qualifications do the authors have?
- Is the source peer-reviewed or has a bias that might influence the findings?
- Is the information provided referenced and are the sources of this information reliable?
- When was the research reported? This is important as theories and concepts can be influenced by the year in which they were published.
- Are the findings reported in the publication supported by other sources? This should be found in the introduction, where the author identifies background information and gaps in the current understanding of the topic.

Table 1.16.1 outlines an example of evaluating a resource step-by-step, for the research article (Figure 1.16.1) that relates to silica aerogels.

TABLE 1.16.1 Evaluation of a resource

Source evaluated: Effect of silica aerogel on thermal insulation and acoustic absorption of geopolymer foam composites: The role of aerogel particle size			
Criteria		**Decision**	**Support/justification**
primary or secondary	Is this an eyewitness account or a second-hand source?	primary	• Research article published in www.ScienceDirect.com • Research results published in article
validity	Does it contain information that is specifically related to the claim?	yes	• Outlines information directly related to the performance of aerogel as thermal insulation.
	Is the evidence and information pertinent to the variables in the research question?	yes	• Information, experiment and results are specific to the performance and physiochemical properties of aerogel.
reliability	Is it current/recent information?	yes	• Published in August 2022 • This is quite recent, but aerogels are modern materials undergoing a considerable amount of research. There may be newer information available.
	Is it up-to-date in its understanding of relationships?	yes	• Information outlined about thermal conductivity and properties of aerogel.
	Is the evidence equivalent to other sources?	mostly	• Table 1 within the article contains a list of six similar experiments.
	Check the credibility—consider who the author is, their qualifications and expertise.	good	• Authors are from the School of Civil Engineering at Wuhan University or the Department of the Built Environment at Eindhoven University. • *Composites Part B: Engineering* is a reputable peer-reviewed journal.
	Try to find the sample size.	known	• Six samples, three trials per sample.
	Try to establish what variables were controlled or measured.	known	• Methodology of the preparation, storage, and testing of the samples is detailed, including controlled variables such as air temperature and humidity.

Effect of silica aerogel on thermal insulation and acoustic absorption of geopolymer foam composites: The role of aerogel particle size

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ABSTRACT

Silica aerogel features super thermal insulating and excellent sound absorption. Although the application of silica aerogel in building materials for energy saving has been investigated recently, the role of aerogel particle size on geopolymer foam render can be crucial but is rarely studied. In this research, four kinds of silica aerogel with different particle sizes (2–40 μm, 100–700 μm, 100–1200 μm, 700–4000 μm) are applied in geopolymer foam aerogel render (GFAR) to improve the thermal insulation and acoustic absorption performance. Results show that larger silica aerogel particles are more beneficial for acoustic absorption and thermal insulation while smaller aerogel particles are less effective for insulation properties. The microstructure of GFAR is investigated in-depth with micro-CT, optical microscopy and ultrasonic pulse velocity test. The mechanism of the improved performance of GFAR with different silica aerogel is explained. The optimal geopolymer foam aerogel render (GFAR) reaches a thermal conductivity of 0.133 W/(m·K), an average value of acoustic absorption coefficient of 0.51 with a density of 715.2 kg/m^3. The result of this study is essential to enlighten the use of larger silica aerogel particles in geopolymer foam render, focusing on a low dosage of silica aerogel while utilize its full benefit.

1. Introduction

The energy-saving of the building sector and the comfort of the indoor living environment have received rising attention due to the increasing awareness of sustainability and the quality of life [1]. The building industry is responsible for almost 40% of the total energy and CO_2 emission in developed countries [2]. For now, many countries have already initiated policies to decrease heat loss through building shells [3]. Moreover, the functionalization of insulation materials is also very important, for instance, improving the acoustic absorption performance to increase the quietness of the indoor environment [4,5] and reduce the noise pollution generated from daily life [6]. Therefore, developing newly high-performance insulating lightweight materials is of significant urgency.

Silica aerogel is a super-insulating inorganic material [7], consisting of a 3D network of crosslinking silica nanoparticles and 95%–99% of air [8]. Thanks to the high porosity and low solid thermal conductivity (λ), the effective thermal conductivity of silica aerogel is pretty low, normally achieving 0.012–0.018 W/(m·K) [9]. However, the silica aerogel is very brittle and easy to break, the commercial silica aerogels are always produced in a powder or particle form [10]. It is often incorporated in other matrices, like fibreglass mats or other lightweight materials to facilitate its benefits [11]. Several investigations related to the incorporation of silica aerogel in building materials have already been carried out as presented in Table 1. Ng et al. [12] prepared high-performance aerogel concrete, with bulk density and thermal conductivity of 1300 kg/m^3 and 0.4–0.5 W/(m·K), respectively. The concrete has a low water to binder ratio of 0.20–0.25 and a silica aerogel replacement ratio of 50%–70% with sand. Tao et al. [13] developed normal cement mortar with an aerogel replacement ratio of 60%, featuring a compressive strength of 8.3 MPa and thermal conductivity of 0.26 W/(m·K). Júlio et al. [14] prepared lightweight aerogel aggregate concrete with a 100% replacement ratio of silica aerogel with sand, resulting in concrete with very low thermal conductivity of 0.085 W/(m·K) but compressive strength of only 0.41 MPa. Seo et al. [15] investigated the silica aerogel used in fly ash/slag geopolymer concrete, with an aerogel concentration of 25%–75%. However, the thermal conductivity is relatively high, reaching 0.8 W/(m·K). Zhu et al. [16] investigated the cement-based mortars and coatings with the addition of silica aerogel. The thermal conductivity is low (0.17 W/(m·K)) with a

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1359-8368/© 2022 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

FIGURE 1.16.1 A peer-reviewed research article entitled Effect of silica aerogel on thermal insulation and acoustic absorption of geopolymer foam composites: The role of aerogel particle size.

Extrapolation to the claim

The first stage of this assessment task involved narrowing the claim to a very specific research question. This could have meant selecting a specific material or different variables. The Extrapolating criterion requires inferring how the trends and patterns identified and explained in the earlier sections of the research report could be extended to the original claim.

When exploring the claim, the dependent and independent variables associated with the claim were identified. Questions that can help extrapolate the findings and conclusion to the claim include:

- What are the dependent, independent and controlled variables associated with the claim?
- What is the impact on the dependent variable when the independent or controlled variables from the research question are extended to include the claim?

Improvements and extensions

Suggest improvements and extension(s) that would complement the research findings and have the potential to provide new evidence that could be used to evaluate the research question (improvements) and claim (extensions) further. These should be linked to the identified limitations (analysis and quality of evidence, being sure not to repeat yourself – you can just identify the issue).

The highest performance level requires that suggested improvements need to be considered and relevant. There are two obvious areas that could be the focus of the suggested improvements.

- A good place to start when proposing possible improvements is the limitations in the research and evidence that were identified and explored in the previous sections in the report.
- What information is required to provide more definitive proof of the research question?

As discussed in the limitation, these could come from the methodology or the interpretation of the results. It could be useful to consider the following:

- changes to the method of selecting samples to reduce bias
- increasing the sample size to increase the accuracy of the results or reduce the errors associated with the measurements
- methods/instruments/techniques used to collect and test the data. If the measurements were below the limit of detection or so low that the associated error was large. Has technology advanced since the previous research to mean that the relationship can be more accurately determined.
- through your investigation what gaps in the evidence did you identify.

These considerations could form the basis for your suggested improvements. While a detailed explanation of these is not required, the report should identify how they would benefit your investigation.

Suggestions of possible extensions to the research investigation could be identified from the information collected as part of the research investigation. Students should ask themselves if there were limits to the scope of evidence available.

- Did the research you found identify areas that could be explored further?
- Could the conclusions from your report be extended to another situation?
- When extrapolating the research question to the claim were there aspects that could not be supported because the information was not available?

PUTTING IT ALL TOGETHER

The sample research investigation report that follows covers the ISMG criteria and may be used as a reference tool. Use this as a model for reference when writing your own report.

Evaluating

Limitations	Relevance	Improvement	Justification
The data does not compare penicillin antibiotics with non-penicillin antibiotics.	The impact of overuse of non-penicillin antibiotics was not explored. This limits reliability of responding to the claim that the world is running out of effective antibiotic treatments.	Research other forms of antibiotics and related diseases, then compare data with the penicillin-based antibiotics and their related diseases.	• The improvement would support the claim and respond to the research question with increased reliability by identifying that antibiotic resistance is present for all types and is a result of over-prescription. • It is valid as it still justifies the claim is true that we are running out of effective antibiotic treatments.
In Evidence 1, the first pneumonia study was conducted on older males over two decades ago with the methodology now outdated. The study in India was impacted by COVID-19 social isolation, had a small sample size and was only conducted over four years.	It does not represent today's data and has both age and gender bias. The mean age was 61.58 years old. The elderly have a higher susceptibility to disease so the mortality rate due to antibiotic resistance is higher, reducing reliability. Small sample size and compounding variable reduces reliability.	Reconduct the data collection and properly examine antibiotic resistance amongst all ages and genders to gather more reliable data. Conduct a follow-up investigation over a longer time period.	• The improvements would increase reliability by compensating for outdated study methodology and increasing the range of sample ages to create a dependable average age that accurately represents a wider population. • This limitation doesn't negate the results as it still demonstrates the claim to be true for the trial group and successfully answers the research question. • To improve reliability by extending the study and conducting it without the compounding variable of COVID-19.
The meningococcal study in Evidence 2 had limitations of the sample area exclusively in England. The data will only reflect a limited population with similar lifestyles in a constrained location.	A restricted number of people able to participate in genome sequencing and antibiotic susceptibility testing. It does not reflect the global population as resistance may be greater in a first world country like England, where antibiotics are widely available.	Increase reliability by expanding data collection. Collecting data globally would allow comparison between countries with high and low antibiotic use and how this affects resistance in their citizens.	• These improvements could be made to increase the accuracy and reliability of data for antibiotic resistance at a global level.
The Russian gonorrhoea study in Evidence 3 did not reveal all information for the samples (gender ratio, age) or test the full range of mutations, resulting in possible data bias. Comparisons between locations were not analysed. In the Bhutan study, cultural, economic and restricted medical facilities plus lack of follow up communication, were severe limitations. The testing method lacked some credibility.	The information from Russia provided by Evidence 3 was lacking personal data. 522 patients were tested from 2015–17. Ethical constraints meant that personal background records were not collected as this is a sexually transmitted disease.	Improvements include more information about the sample group to judge if it is a fair test or contains bias. Comparative analysis between locations of data collection to indicate if resistance may be the result of an outside element instead of purely penicillin-based resistance.	The suggestions would improve the validity and reliability of the data by allowing for judgement on potential bias.

Extensions

The extensions that could be implemented for future studies include:

Expanding the sample size and collection locations would allow for accurate data regarding the scale of antibiotic resistance amongst global populations and can form a clearer understanding and knowledge of other causes of ineffective antibiotic treatment, outside of overuse.

Investigating the specific mutations and strains of pneumonia, meningococcal and gonorrhoea would compare the resistance rate amongst different strains and any increased occurrence of resistance. Insight

would have been provided for the mutations affecting a person's immune ability to eradicate or resist the disease as well.

Researching a specific antibiotic, rather than a group of similar ones, would allow for clearer comparisons to be made regarding the level of resistance to the chosen disease. By specifying the antibiotic, clearer data would have been provided, increasing the reliability of the results.

Extending the diversity of isolates tested would have decreased the rate of bias. By capturing an equal ratio of genders, examining patients of all ages and including diverse nationalities and lifestyles, it would have better reflected the overall population, therefore indicating a reliable average rate of resistance amongst the global population.

Conclusion

To summarise, bacteria causing pneumonia, meningococcal and gonorrhoea have displayed growing resistance over time against penicillin-based antibiotic treatments. Evidence 1 showed a recorded resistance in pneumonia at 18.5%, while Evidence 2 showed an increase in resistance for meningococcal from 2.8% to 35.88%. Lastly, Evidence 3 showed that gonorrhoea resistance was up at 72.4%. The evidence presented further developed the relationships between resistance, sample age and over-prescription. Sample age and over-prescription of penicillin-based antibiotics have resulted in a greater rate of antibiotic resistance in the bacteria through increased levels of antibiotic exposure over time (Willerton, 2021). This shows conclusive evidence that over-prescription of penicillin-based antibiotic treatments is negatively impacting human ability to eradicate diseases such as pneumonia, meningococcal and gonorrhoea due to increased antibiotic resistance. Combined with other data on the inadequate research and development of new antibiotics, the evidence therefore supports the claim, 'We are running out of effective antibiotic treatments'.

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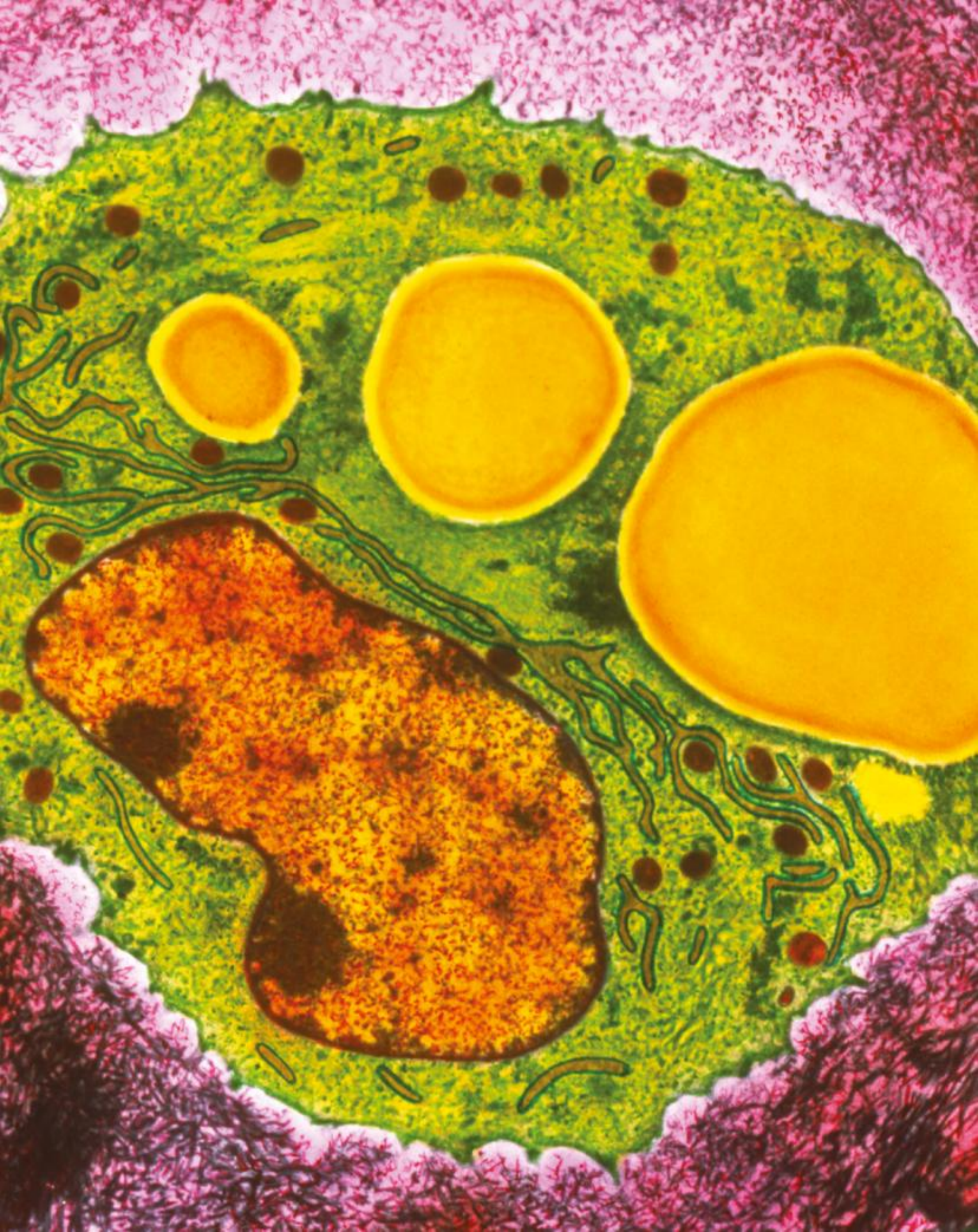

UNIT 1 Cells and multicellular organisms

TOPIC 1 Cells as the basis of life

TOPIC 2 Exchange of nutrients and wastes

Unit 1 objectives

- Describe ideas and findings about cells as the basis of life; exchange of nutrients and wastes; and cellular energy, gas exchange and plant physiology.
- Apply understanding of cells as the basis of life; exchange of nutrients and wastes; and cellular energy, gas exchange and plant physiology.
- Analyse data about cells as the basis of life; exchange of nutrients and wastes; and cellular energy, gas exchange and plant physiology.
- Interpret evidence about cells as the basis of life; exchange of nutrients and wastes; and cellular energy, gas exchange and plant physiology.
- Evaluate processes, claims and conclusions about cells as the basis of life; exchange of nutrients and wastes; and cellular energy, gas exchange and plant physiology.
- Investigate phenomena associated with cells as the basis of life; exchange of nutrients and wastes; and cellular energy, gas exchange and plant physiology.

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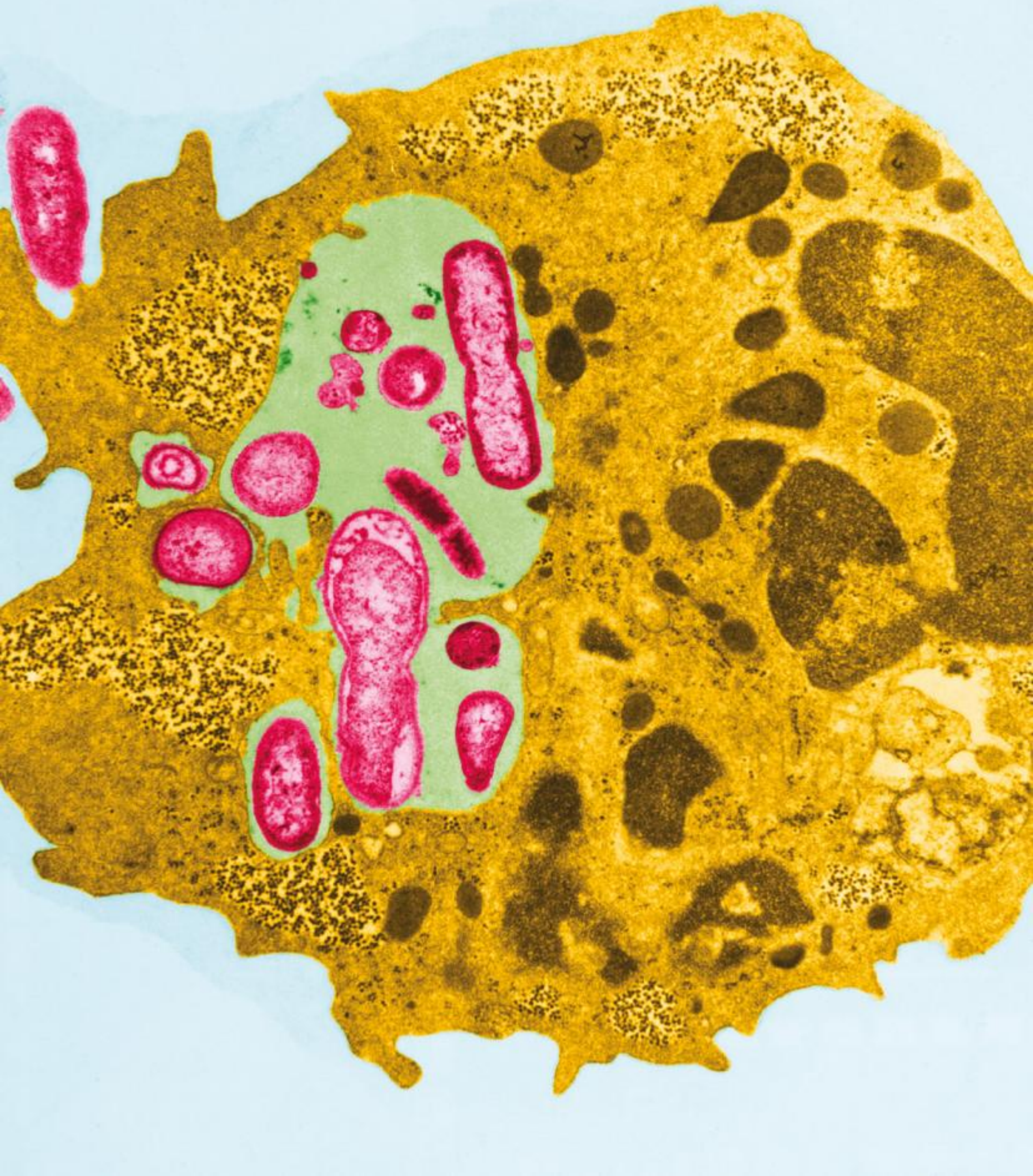
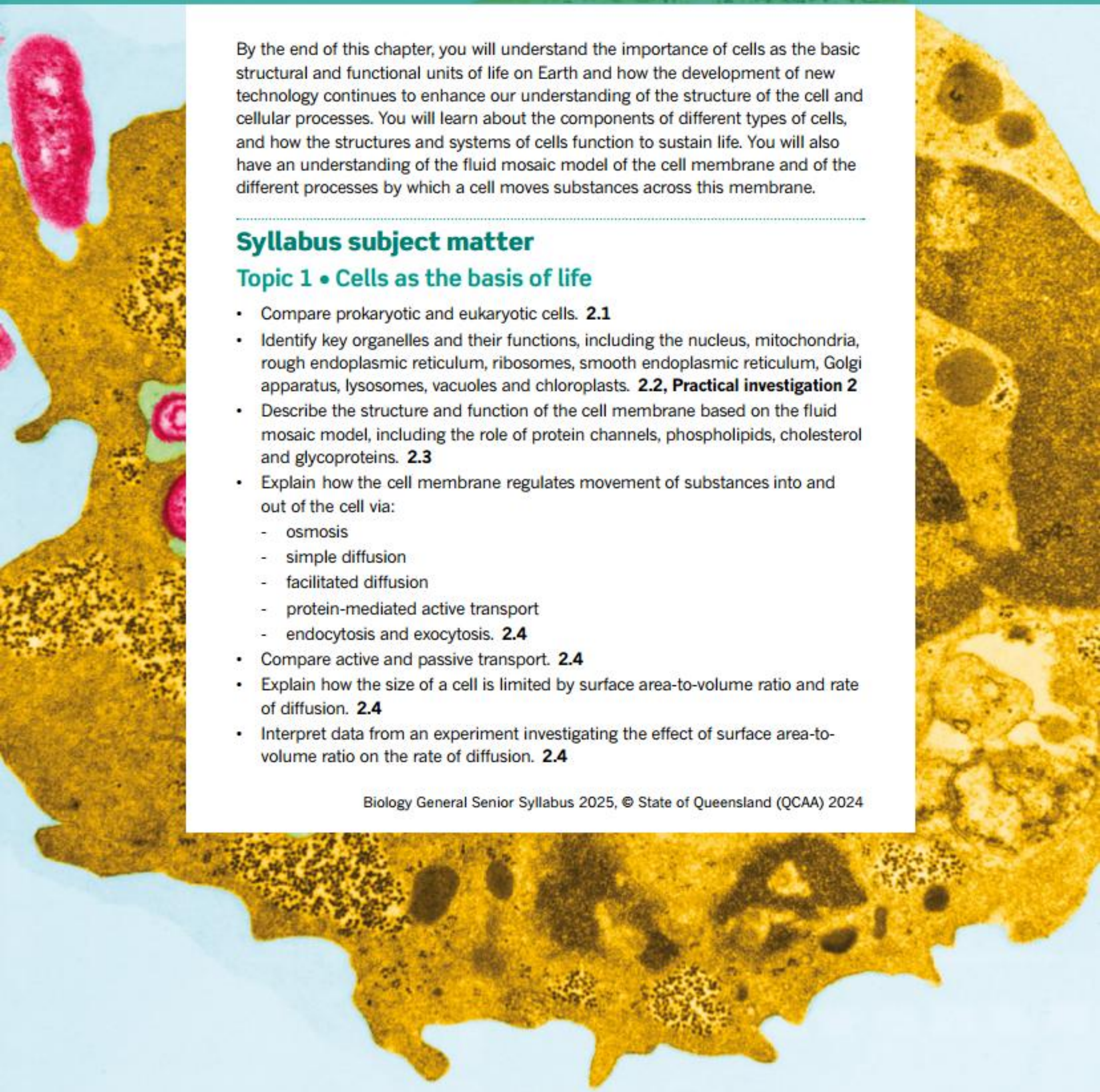


CHAPTER

02 The fundamental cell



By the end of this chapter, you will understand the importance of cells as the basic structural and functional units of life on Earth and how the development of new technology continues to enhance our understanding of the structure of the cell and cellular processes. You will learn about the components of different types of cells, and how the structures and systems of cells function to sustain life. You will also have an understanding of the fluid mosaic model of the cell membrane and of the different processes by which a cell moves substances across this membrane.

Syllabus subject matter

Topic 1 • Cells as the basis of life

- Compare prokaryotic and eukaryotic cells. **2.1**
- Identify key organelles and their functions, including the nucleus, mitochondria, rough endoplasmic reticulum, ribosomes, smooth endoplasmic reticulum, Golgi apparatus, lysosomes, vacuoles and chloroplasts. **2.2, Practical investigation 2**
- Describe the structure and function of the cell membrane based on the fluid mosaic model, including the role of protein channels, phospholipids, cholesterol and glycoproteins. **2.3**
- Explain how the cell membrane regulates movement of substances into and out of the cell via:
  - osmosis
  - simple diffusion
  - facilitated diffusion
  - protein-mediated active transport
  - endocytosis and exocytosis. **2.4**
- Compare active and passive transport. **2.4**
- Explain how the size of a cell is limited by surface area-to-volume ratio and rate of diffusion. **2.4**
- Interpret data from an experiment investigating the effect of surface area-to-volume ratio on the rate of diffusion. **2.4**

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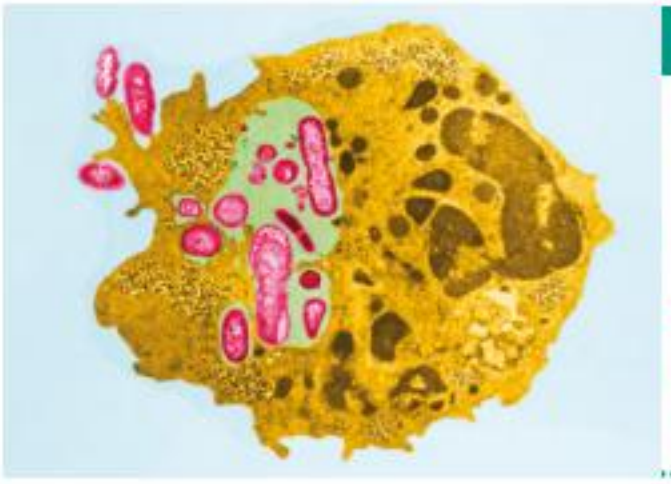
2.1 Cell types



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand the importance of cells as the basic structural and functional units of life on Earth
- understand the differences and similarities between prokaryotes and eukaryotes
- identify key organelles and their functions
- explain the role of the cell membrane
- understand the difference between active and passive transport.

Cells are the basic structural units of all living things. The cell theory is one of the fundamental principles of biology. It is based on microscopic and experimental studies of tissues, from all types of organisms, carried out over the last 300 years.

In this module, you will learn about cell theory, the differences between plant and animal cells, and the microscopy techniques that are used to view cells and their components.

CELL THEORY

The cell theory was developed over hundreds of years by scientists of various nationalities and depended on the technology available at the time.

Cells are the basic structural units of living organisms. The cell theory states that:

- all organisms are composed of cells
- all cells come from pre-existing cells
- the cell is the smallest living organisational unit.

Biogenesis

The cell theory states that all cells arise from pre-existing cells. This is known as **biogenesis**.

Until the 1850s, the idea of spontaneous generation was accepted as the origin of small organisms, such as maggots. According to the theory of spontaneous generation, some organisms could suddenly form from certain types of matter, such as a grain of sand or dead flesh.

However, experiments by Francesco Redi on maggots in the 17th century and Lazzaro Spallanzani on microorganisms in the 18th century refuted spontaneous generation. These scientists showed that the presence of maggots and microorganisms was a result of contamination rather than spontaneous generation.

The first primitive microscopes allowed little detail of a cell's internal structure to be seen but new technologies have resulted in a greater knowledge of the structure and function of cells.

COMMON CELL FEATURES

Although there are different types of cells, the cells of plants, animals and bacteria have a number of common features. These common features include:

- a **cell membrane (also called a plasma membrane)**, which separates the interior of the cell from the outside environment
- **cytoplasm**, which consists of the **cytosol** and specialised structures called **organelles**. Cytosol is a gel-like substance that is made up of more than 80% water and contains ions, salts and organic molecules

- **deoxyribonucleic acid (DNA)**, which carries hereditary information, directs the cell's activities and is passed from generation to generation
- **ribosomes**, which are organelles responsible for the synthesis of **proteins**.

CELL SIZE

Cells vary greatly in size, as can be seen in Figure 2.1.1. Most cells are microscopic and you need a microscope to see them. Cell size is usually measured in micrometres. There are 1000 micrometres (μm) in 1 millimetre (mm). There are exceptions, such as the egg cell of some bird species, which can be many centimetres in diameter. Some typical cell sizes are as follows:

- bacterium: 0.1–5 μm long
- human: 10–200 μm long
- *Paramecium* (a single-celled eukaryote): about 150 μm long.

The thickness of cell membranes also differs and can be 0.004–0.1 μm.

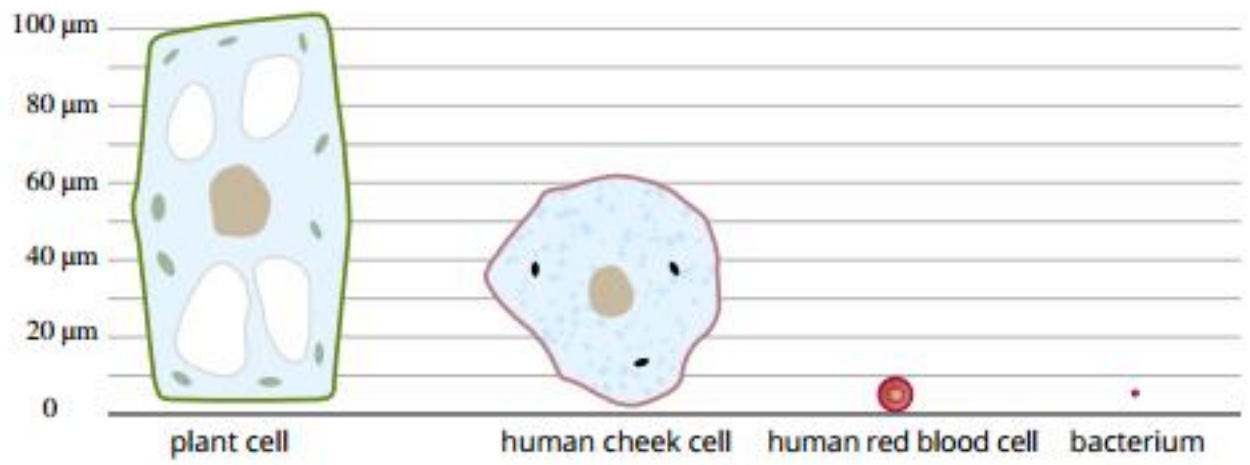


FIGURE 2.1.1 A typical plant cell, human cheek cell, human red blood cell and bacterium. Note the great difference in size between the plant and animal (human) cells and the bacterium.

The development of sophisticated microscopy technology to observe cells and intracellular particles has allowed scientists to measure increasingly smaller objects (Figure 2.1.2). This has led to the development of appropriate units of measurement to describe microscopic lengths.

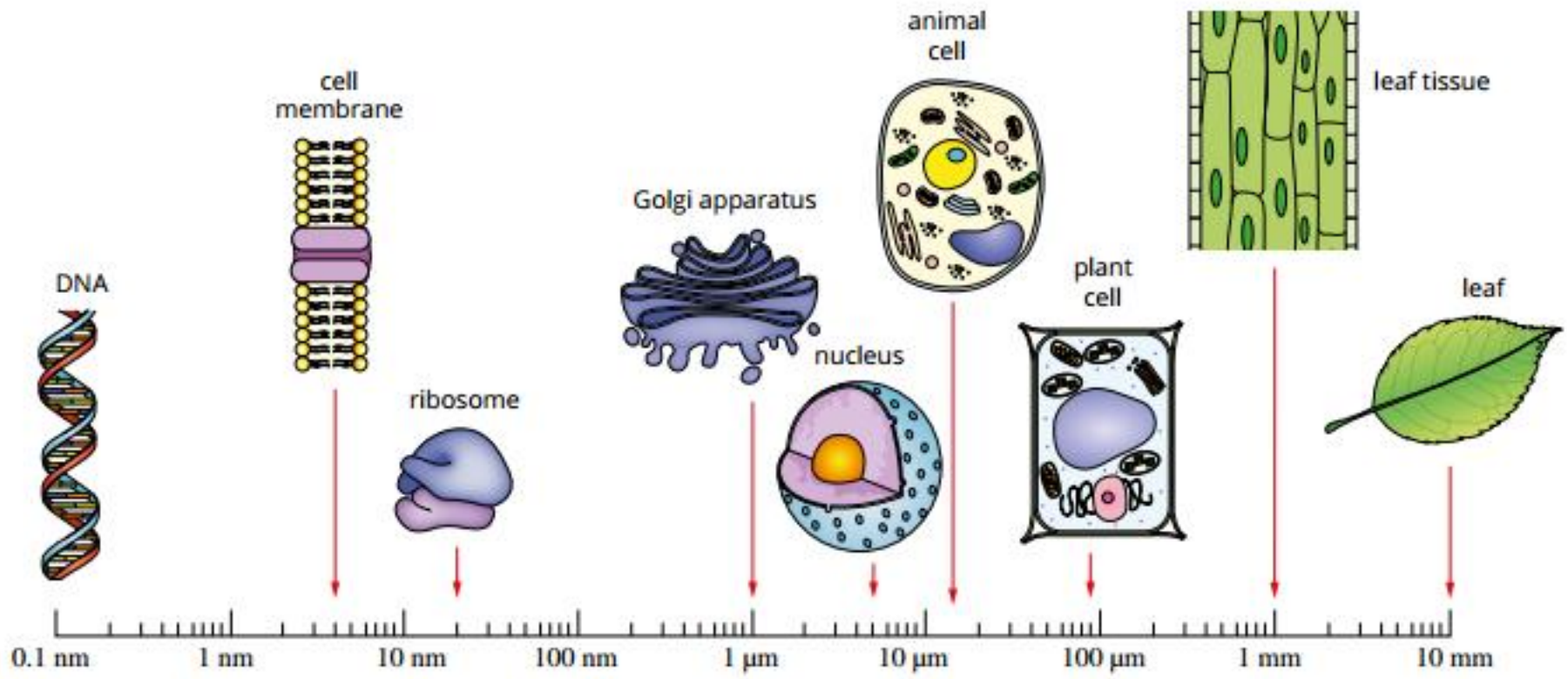


FIGURE 2.1.2 The scale shows the range of size of a variety of cells, organelles and molecules within a cell. The scale is logarithmic to accommodate the range of sizes shown.

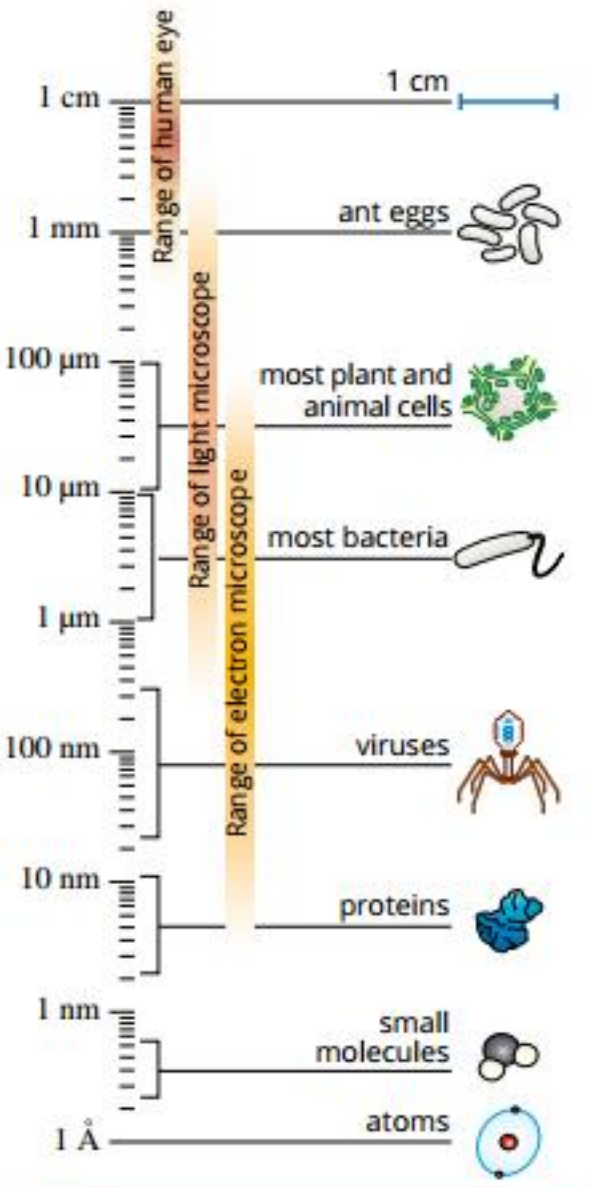


FIGURE 2.1.3 A comparison of the ranges of the light and electron microscopes (the scale is logarithmic)

In the International System of Units (SI units), the unit for length is the metre (m). Table 2.1.1 illustrates the derivation of the smaller units of length in relation to the metre. Refer to 1.4 Units for detailed assistance with units of measurements.

TABLE 2.1.1 SI units for smaller units of length relative to the metre

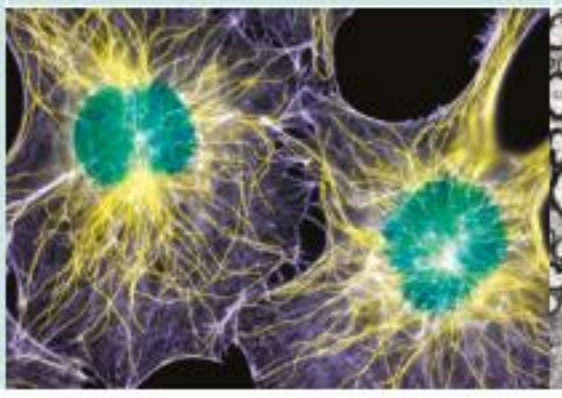
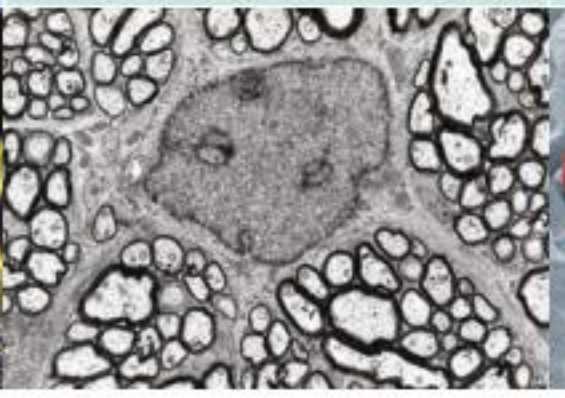
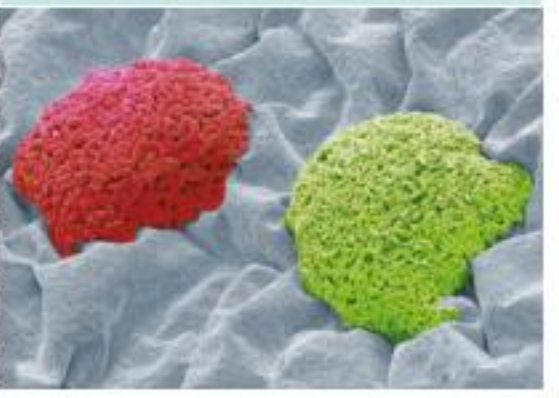
Fraction of a metre	Units	Symbol	
one hundredth = $\frac{1}{100}$ = 0.01 = 10^{-2} m	centimetre	cm	1 m = 10^2 cm
one thousandth = $\frac{1}{1000}$ = 0.001 = 10^{-3} m	millimetre	mm	1 m = 10^3 mm
one millionth = $\frac{1}{1000000}$ = 0.000001 = 10^{-6} m	micrometre	µm	1 m = 10^6 µm
one thousand millionth = $\frac{1}{1000000000}$ = 0.000000001 = 10^{-9} m	nanometre	nm	1 m = 10^9 nm

INVESTIGATING CELLS

Cytology is the study of cells. Cytologists (scientists who study cell structure and function) and histologists (scientists who study tissue structure) use a variety of tools and techniques, including several microscopy techniques. Modern microscopy techniques, including light and electron microscopy (Table 2.1.2), have greatly advanced our understanding of the structure and function of cells. The type of microscope used depends on the characteristics and properties of the specimen to be observed, such as the size of cell or cell component and whether it is living or dead (Figure 2.1.3). Scientists also consider the access to and costs of using specialist facilities and preparing specimens when deciding which microscopy technique to use.

TABLE 2.1.2 A comparison of modern microscopy techniques

	Light microscope	Transmission electron microscope	Scanning electron microscope
Radiation source	light	electrons	electrons
Wavelength (nm)	400–700	0.005	0.005
Lenses	glass	electromagnetic	electromagnetic
Specimen	living or non-living supported on glass slide	non-living supported on a small copper grid in a vacuum	non-living supported on a metal disc within a vacuum
Maximum resolution (nm)	200	1	10
Maximum magnification	1500×	250000×	100000×
Stains	coloured dyes	impregnated with heavy metals	coated with carbon or gold
Type of image	may be coloured	monochrome unless stained	monochrome unless stained

Light microscopy

Most cells are so small that they can only be seen with a microscope like the one shown in Figure 2.1.4. The light microscope uses light and a system of lenses to magnify the image. One lens is called the objective lens, and the other is the eyepiece or ocular lens. Most school laboratories use the compound light microscope. Other types of light microscopes are the fluorescent light microscope, which can be used to see materials that fluoresce naturally, or which have been stained with a fluorescent dye and the confocal microscope that can produce highly detailed three-dimensional pictures of microscopic structures.

One of the main advantages of light microscopy is that it allows you to view living cells in colour.

Sample preparation is usually quick and simple. Stains can be used to highlight different components of cells in colour. A thin specimen is mounted on a glass slide and placed on the stage under the lenses. Light travels through the specimen and into the lens system, and the image is viewed by eye or with a digital camera (Figure 2.1.5).

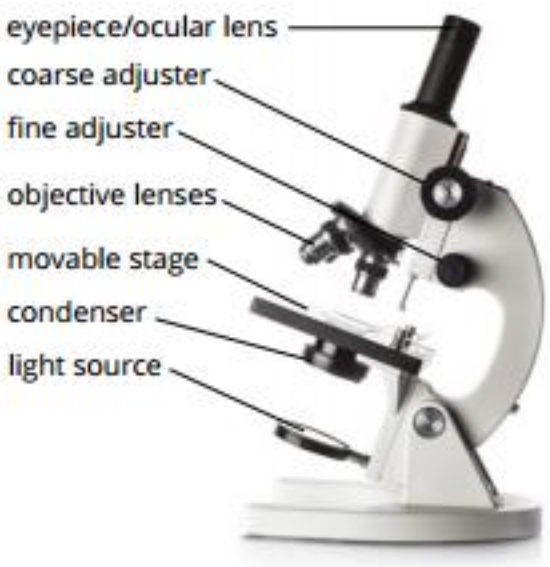


FIGURE 2.1.4 A light microscope and its parts

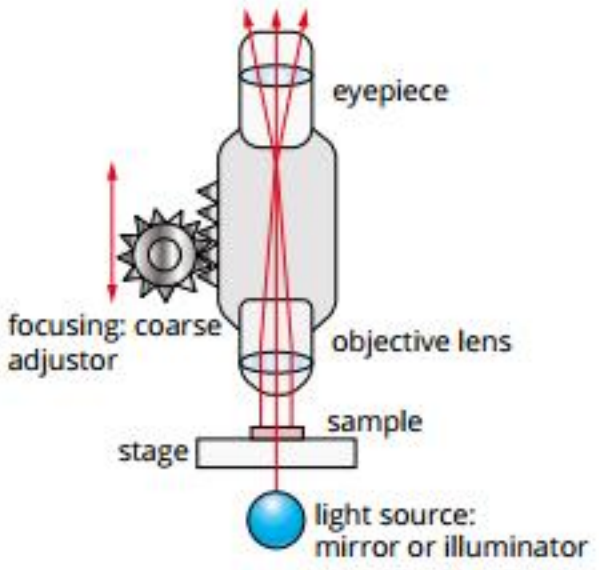


FIGURE 2.1.5 The lenses of a light microscope magnify the image.

The condenser lens beneath the movable stage concentrates light from the light source onto the specimen, and the image is focused by the coarse and fine adjusters. Different parts of the specimen can be viewed by moving the specimen on the stage.

Light microscopy techniques used in cytology include histology, autoradiography, fluorescence and confocal microscopy. Each of these uses visible light to examine cells and tissues.

Electron microscopy

In electron microscopy, an object is viewed by using an electron beam instead of light. This allows structures to be seen in far more detail than is possible with light microscopy. An electron microscope produces a narrow beam of electrons that is maintained by electromagnetic lenses, which are coils that surround the tube and emit an electromagnetic field. Electrons striking the specimen are absorbed, scattered or pass through it. The image is then recorded digitally and processed.

The image obtained with an electron microscope has a much higher resolution and greater depth of field than an image from a light microscope. Electron microscopy produces only black and white images, but these are often coloured later to highlight important features.

Transmission electron microscopy

Figure 2.1.6 shows a transmission electron microscope. In transmission electron microscopy (TEM), the electron beam travels through an ultrathin section (less than 100 nm thick) of a specimen. This allows very fine details of cellular structures to be seen, like those shown in Figure 2.1.7.

FIGURE 2.1.6 A transmission electron microscope. Specimens are specially prepared, and the image is taken within a vacuum to ensure the electron beam remains focused. Therefore, only non-living materials can be observed under the transmission electron microscope.

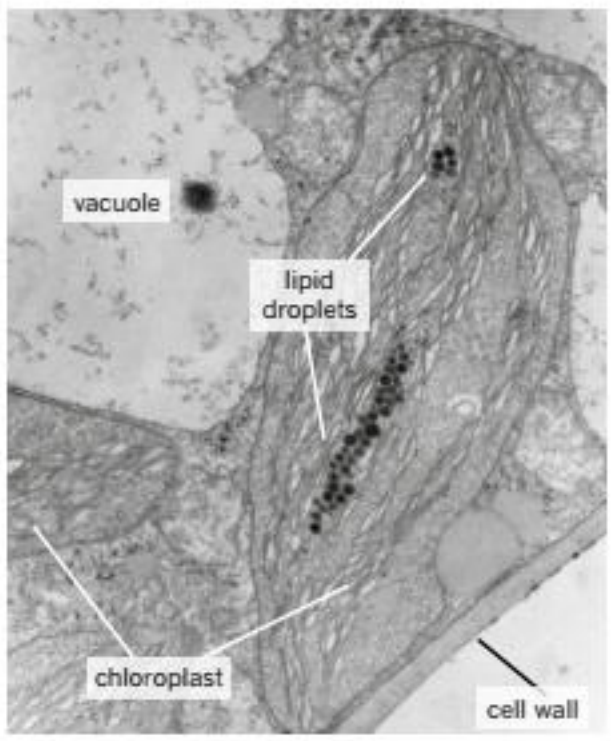


FIGURE 2.1.7 A transmission electron micrograph of a plant leaf, showing chloroplasts, with lipid droplets showing in one. The pale area to the upper left is a vacuole and the cell wall is also visible at the lower right.

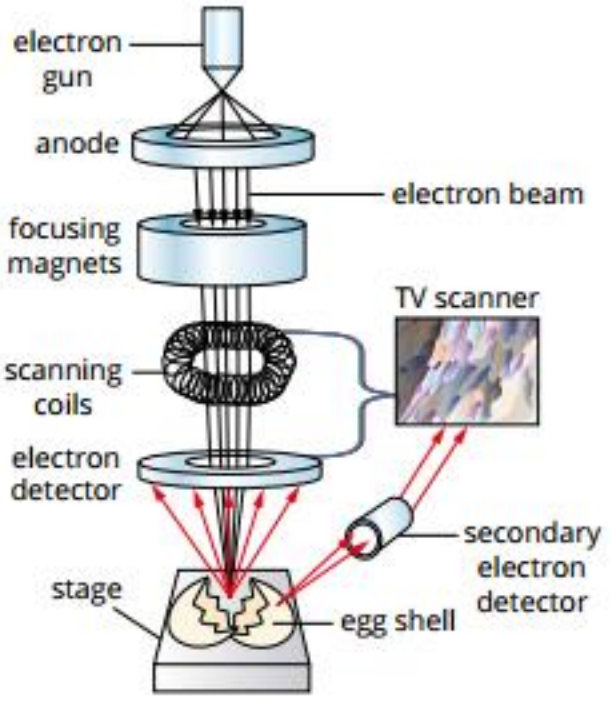


FIGURE 2.1.8 The scanning electron microscope detects secondary electrons emitted by atoms excited by the electron beam. The specimen is prepared by coating it with an electron-dense, electrically conductive material such as gold and therefore can only be used on non-living objects.

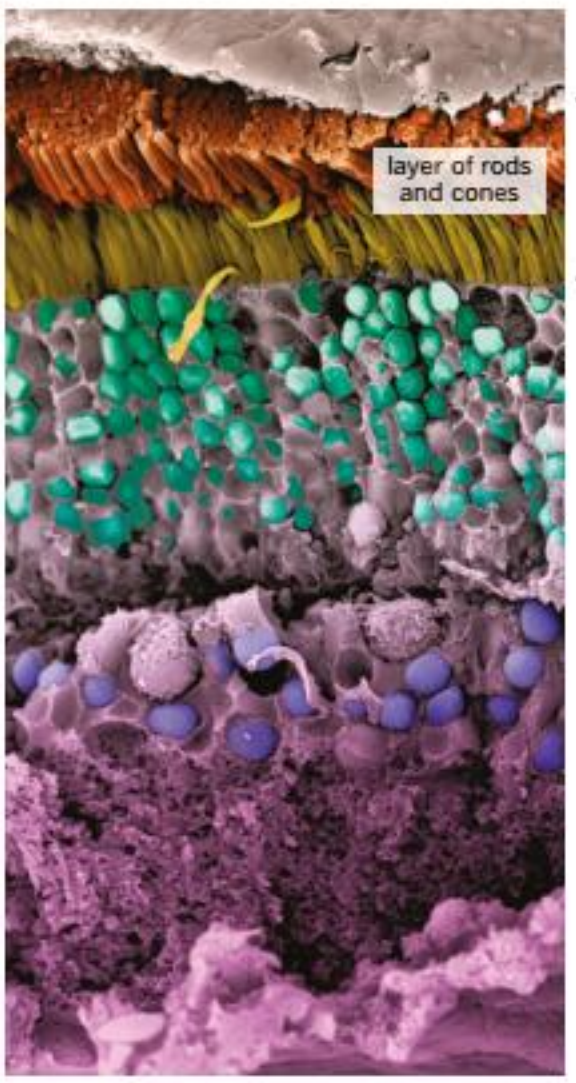


FIGURE 2.1.9 A coloured scanning electron micrograph of a section through the retina of an eye, showing cone and rod cells

Because the specimen must be in a vacuum in the transmission electron microscope, the specimen is first chemically fixed to stop the structures from collapsing and then dehydrated with alcohol. It is then embedded in a plastic resin, sectioned with a diamond cutter called an ultramicrotome, and stained.

Scanning electron microscopy

Figure 2.1.8 shows a scanning electron microscope. In scanning electron microscopy (SEM), the electrons bounce off a specimen that has been coated with an extremely thin layer of an electrically conducting material such as gold. This gives a high-resolution image of the surface features but cannot show internal details (Figure 2.1.9).

Autoradiography

Autoradiography is a method that allows scientists to identify specific organelles or the location of molecules within a cell or tissue. The tissue is first treated with a radioactively labelled substance that is taken up into the part of the cell that is being investigated, such as the nerve tissue shown in Figure 2.1.10. The tissue is sliced into very thin sections that are placed against a very thin high-resolution photographic film. The radioactive substance emits beta particles that produce an image on the film. The tissue sections are stained so that the photographic image can be located in relation to cellular structures. This technique can be used to indicate the organelles are active under particular circumstances.

Although autoradiography is still sometimes used with light microscopy, it is more commonly used with electron microscopy.

CELL CLASSIFICATION

As the study of cells advanced it became clear that cells show one of two patterns; simple cells, called **prokaryotic cells**, and more complex cells, called **eukaryotic cells**. In earlier classification systems, all organisms were divided into five ranks, called kingdoms. Prokaryotic organisms were placed in the kingdom Monera, and eukaryotic organisms were placed in the kingdoms: Protista, Plantae, Fungi and Animalia. These systems were based on the morphology (appearance and structure) of the organisms and their cells.

FIGURE 2.1.10 An autoradiograph of a slice through nerve tissue from the visual centre of the brain, showing how visual messages from one eye are received by the brain. Rows of neuron (nerve cell) cubes are laid out in columns on the outside of the brain tissue, and the active areas of the brain have absorbed a radioactive chemical. The glow is developed onto photographic paper and produces an autoradiograph.

In the late 1970s, the use of **DNA (deoxyribonucleic acid)** techniques in the emerging field of evolutionary genetics led to the discovery of two different types of prokaryotic cells. This resulted in the development of a system used today, with three domains and six kingdoms (Figure 2.1.11). Domains are now the highest rank in **taxonomy**, instead of kingdoms. Prokaryotes are divided into two domains: Bacteria and Archaea. All eukaryotic organisms are placed in a third domain called Eukarya. The four kingdoms within the Eukarya domain remain the same: Protista, Plantae, Fungi and Animalia.

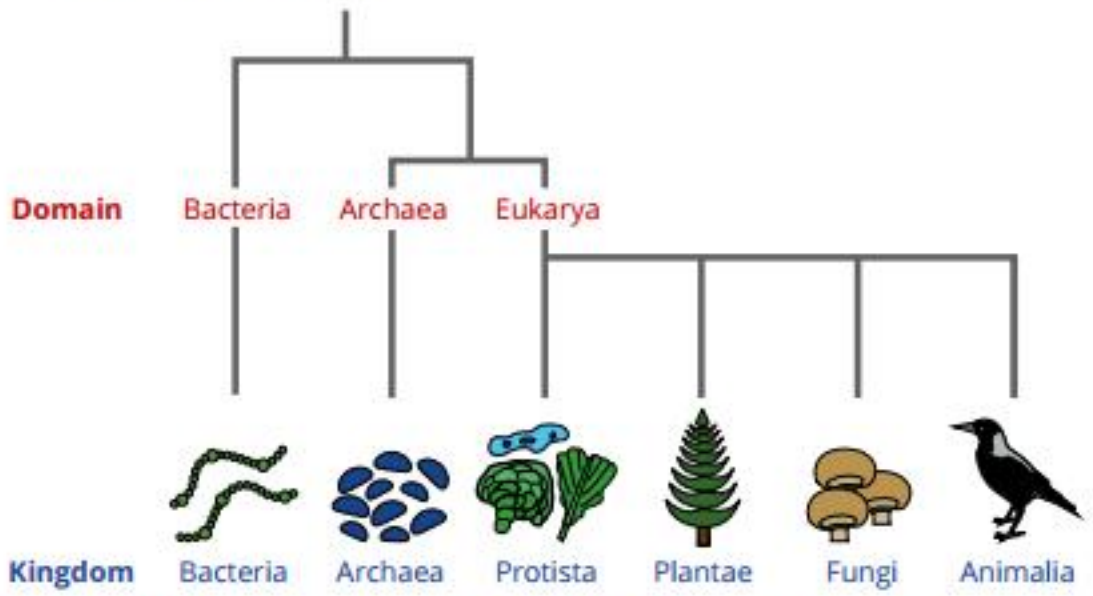


FIGURE 2.1.11 The classification of living things, showing the three domains based on cell types, and the six kingdoms. Bacteria and Archaea have prokaryotic cells. Protista, Plantae, Fungi and Animalia have eukaryotic cells.

PROKARYOTIC CELLS

Prokaryotic organisms are unicellular and have a simple cell structure. **Bacteria**, cyanobacteria (photosynthetic bacteria) and **archaea**, such as methanogens, are examples of prokaryotes. Prokaryotic organisms can be found everywhere, even in extreme environments such as volcanoes.

Most prokaryotic cells are small and therefore have a large surface area relative to their volume (see Module 2.4 for a discussion on surface area-to-volume ratio). This allows the cells to take in and release materials efficiently and replicate quickly.

The structure of a typical prokaryotic cell is shown in Figure 2.1.12. Prokaryote cells lack membrane-bound organelles, and their cytoplasm contains scattered ribosomes that are involved in the synthesis of proteins. The genetic material of prokaryotic cells is usually a single, circular DNA **chromosome** called a **genophore**. The genophore is contained in an irregularly shaped region called the **nucleoid**. Unlike the nucleus of eukaryotes, the nucleoid does not have a nuclear membrane.

The chromosomal DNA of prokaryote cells is attached to the cell membranes by a region of the chromosome called the origin. In addition to this chromosomal DNA, many prokaryotic cells also contain small rings of double-stranded DNA called **plasmids**.

The cell membrane of prokaryotic cells is surrounded by an outer **cell wall**. Many bacteria also have a capsule outside the cell wall. The capsule protects the cell from damage and dehydration.

Many prokaryotes have flagella that enable them to move freely. Some also have small hair-like projections called pili, which are involved in the transfer of DNA between organisms and help movement. Specialised pili that can attach to surfaces are called fimbriae.

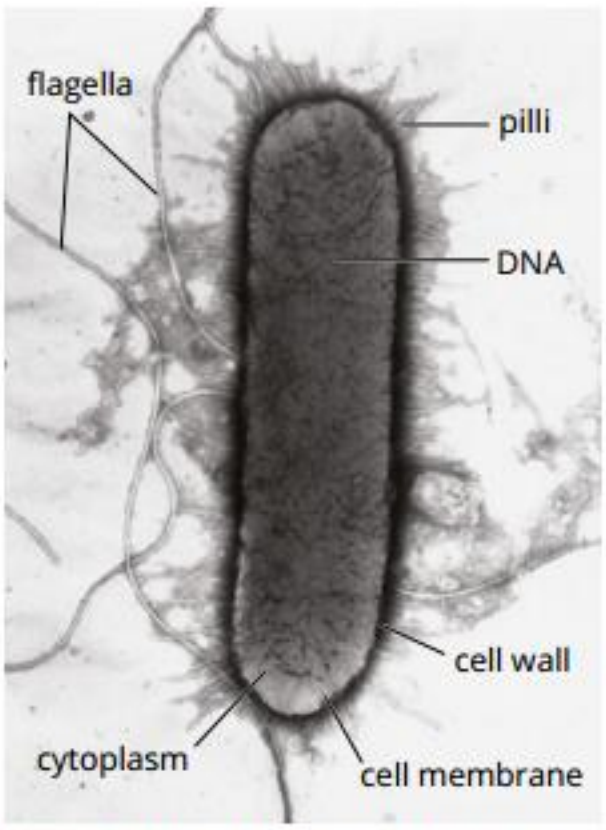


FIGURE 2.1.12 A typical prokaryotic bacterial cell

Bacteria

Most prokaryotes in the domain Bacteria are microscopic single-celled organisms. Fossil evidence dated at 3.5 billion years old confirms that bacteria were the first type of living organisms on Earth. Today, bacteria are still the most numerous type of organism in the biosphere.

Bacteria have very diverse metabolisms and can survive in a great range of habitats and conditions. For example, numerous species of bacteria are common in environments of moderate temperature that are moist and low in salt, where sunlight or **organic compounds** are plentiful, and in or on plants and animals.

Other species of bacteria need little oxygen to survive because they have evolved specialised chemical pathways to extract energy from their environment and manufacture complex energy-rich molecules such as carbohydrates (see Module 3.1). Bacteria can obtain energy from sunlight (photosynthesis) or by reducing **inorganic compounds** such as sulfates or ferric ions (chemosynthesis).

Bacteria play an important role in ecosystems because they break down many kinds of substances, including plant and animal remains and wastes. Bacteria are also widely used in industry to manufacture foods such as cheeses and yoghurt, and in medicine, to produce antibiotics, drugs and even human insulin. Some bacteria can break down oils and plastics, which makes them useful for pollution control.

Carbohydrates are organic compounds of carbon, hydrogen and oxygen, with the number of hydrogen and oxygen atoms in the ratio 2:1. This ratio of 2:1 is the same ratio of hydrogen to oxygen for water. Sugars and starches are examples of carbohydrates. Proteins and carbohydrates molecules sometimes combine as complex structures (peptidoglycans) to become part of the cell wall of bacteria.

Archaea

The prokaryotes in the domain Archaea include **extremophiles**. These are organisms that can live in extreme conditions, such as:

- areas of high temperatures (thermophiles)
- areas of low temperatures
- the upper atmosphere
- very alkaline environments
- very acidic environments (acidophiles)
- very salty environments (halophiles)
- environments with little or no oxygen
- areas without light
- petroleum deposits deep underground.

Archaea hold records for living in the hottest places (121°C), the most acidic environments (pH 0), and the saltiest water (about 30% salt). However, some archaea live in less extreme environments, such as open seas.

The unique place of archaea among living organisms was not recognised for a long time. The main reason for this was because the extreme habitats where they live made it difficult for scientists to find archaea organisms and to culture them in a laboratory. Another reason is that most archaea look very similar to bacteria, even though they are as different from bacteria as humans are.

The ability of archaea to live in extreme environments is partly due to their unique cell membranes. Like other living organisms, archaea possess a cell membrane composed mainly of lipids. Cell membranes need to be fluid to respond to external deformations and damage and allow proteins to move around.

The lipids in eukaryotic cell membranes have fluidity and selective permeability, but only in a narrow range of temperatures and pressure. The lipids in archaean membranes are different because they form a unique cell membrane structure. The structure remains fluid and permeable over a wide range of temperatures, from freezing cold to boiling hot, and at extreme depths of the ocean floor.

There are many different types of extremophiles. Hyperthermophiles such as *Pyrococcus furiosus*, which is shown in Figure 2.1.13, can survive in very hot environments such as undersea vents, where temperatures are often above 100°C. *Pyrococcus* can also thrive under high pressure—they are barophilic. This means they can withstand the extremely high pressure at the ocean floor. *Sulfolobus* species, which live in volcanic springs, are thermophiles as well as acidophiles—they can survive both high temperatures and high acidity.

Lipids are 'fatty' organic compounds, including fats and oils, composed mainly of carbon, hydrogen and oxygen. Lipids have proportionally less oxygen than carbohydrates, and may contain other elements. Lipids are an integral structural component of cell membranes.

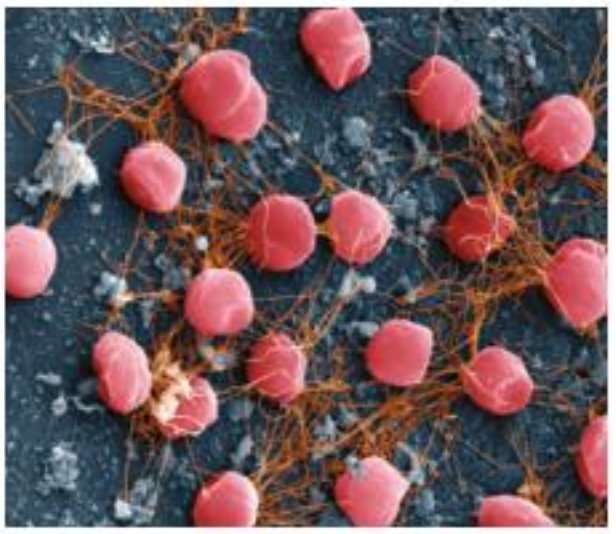


FIGURE 2.1.13 A scanning electron micrograph of hyperthermophile *Pyrococcus furiosus*. *Pyrococcus* can only exist in very hot environments such as hot undersea vents.

Sulfolobus is shown in Figure 2.1.14. Extremophiles have evolved many unique adaptations to ensure their continued existence in environments where most organisms are not able to survive.

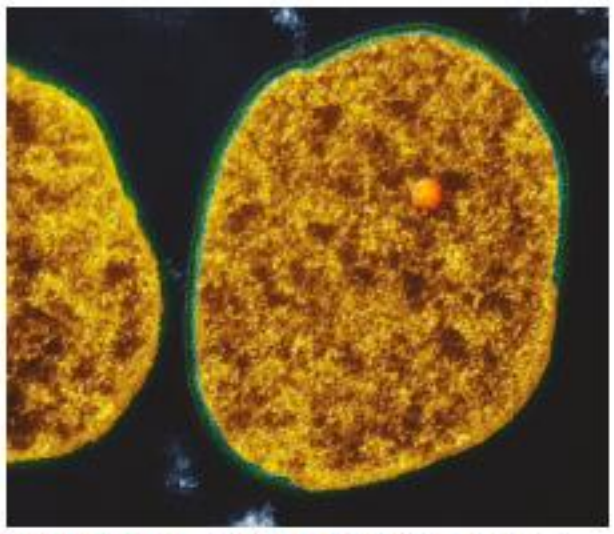


FIGURE 2.1.14 *Sulfolobus* are thermophiles as well as acidophiles. They thrive in hot, acidic environments.

Differences between bacteria and archaea

Despite their name, archaea are not the most ancient group of organisms. DNA studies have shown that bacteria are the most ancient group. The evolutionary relationship between archaea, eukaryotes and bacteria remains unclear. While archaea and gram-positive bacteria share many structural features and metabolic pathways, suggesting a common ancestor, many other archaea genes are more similar to the genes found in eukaryotic cells.

The cells of bacteria and archaea are different in a number of ways.

- Archaea have a different type of lipid structure in the cell membranes.
- Bacterial cell walls contain **murein**; archaean cell walls do not contain murein (although there is a similar compound in some archaea).
- Both have diverse metabolic systems, but methanogenesis (in which methane is produced) is unique to archaea.

> **Gram staining** is used to classify bacteria into two groups—gram-positive and gram-negative. The stain reacts to chemicals in the cell walls of bacteria and makes the bacteria appear pink/red (gram-positive) or blue/purple (gram-negative).

EUKARYOTIC CELLS

Eukaryotic cells are relatively large and more complex than prokaryotic cells. They possess membrane-bound organelles such as a nucleus and mitochondria. Eukaryotic cells are found in multicellular organisms such as protists, fungi, plants and animals. Organisms that are composed of eukaryotic cells are called 'eukaryotes'. A typical plant cell and a typical animal cell is shown in Figure 2.1.15.

As well as a cell membrane surrounding the cytoplasm, eukaryotes have internal membranes that form specialised compartments within the cell. This is known as **cell compartmentalisation**. The membrane-bound compartments are organelles, which are specialised structures that have specific functions. You will learn more about cell compartmentalisation in Module 2.2. As well as compartmentalisation, many eukaryotic cells are specialised, with particular cells undertaking particular functions (e.g. producing hormone or making particular enzymes).

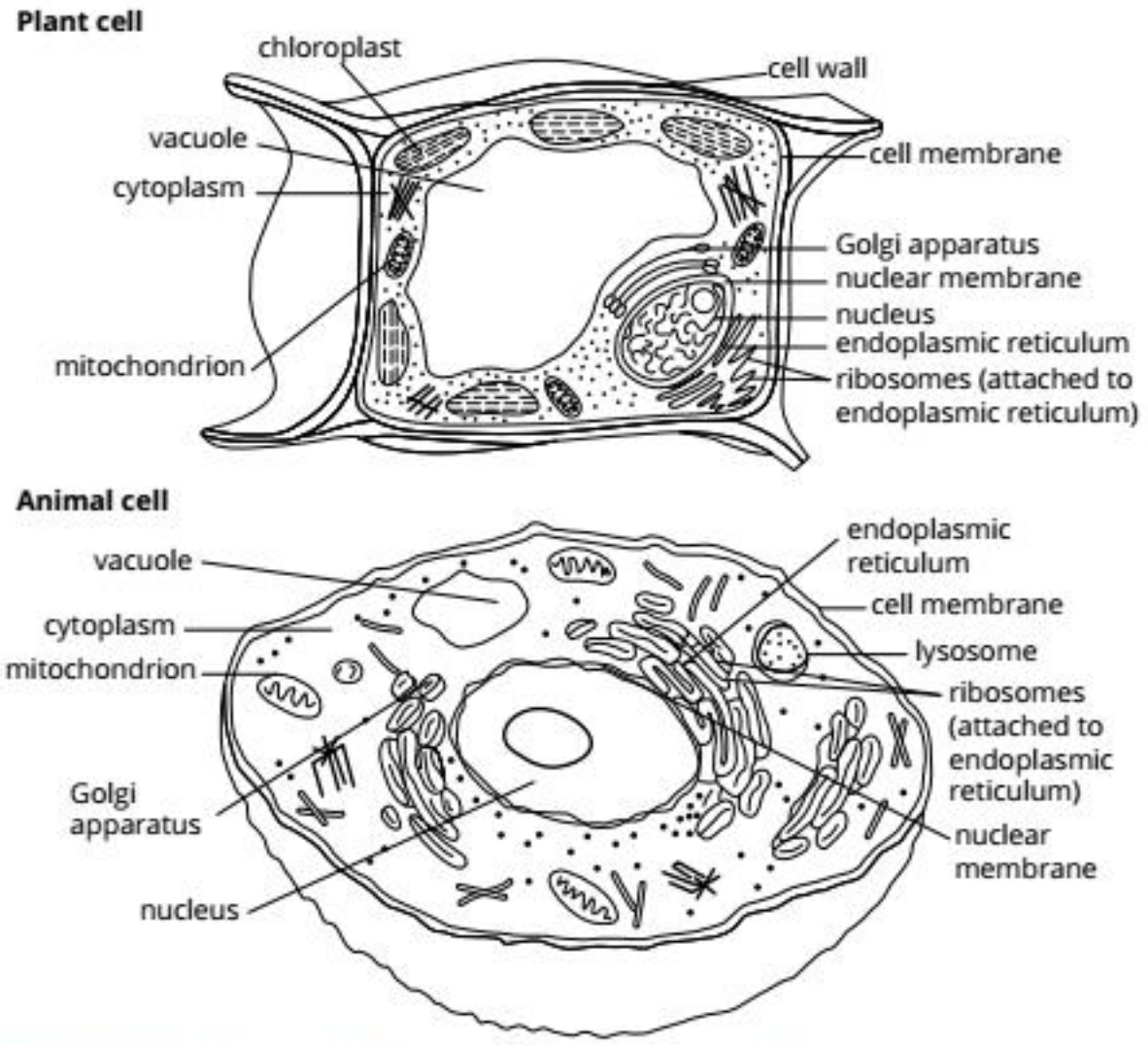


FIGURE 2.1.15 Diagram of a typical plant cell and an animal cell, showing major organelles

Organelles in eukaryotic cells include a nucleus, mitochondria, rough endoplasmic reticulum, smooth endoplasmic reticulum, Golgi apparatus, lysosomes, vacuoles and chloroplasts. These are all membrane-bound organelles and are only found in eukaryotes. Also present in eukaryotic cells are ribosomes that have no membrane and are also found in prokaryotes. You will learn more about these organelles in Module 2.2. Centrioles, which are found in some eukaryotes, also have no membrane. They are involved in assisting cells to divide and will be discussed in Module 4.2. Movement of substances into, out of and around the cell allows the maintenance of basic metabolic processes such as cellular respiration, homeostasis and reproduction. Eukaryotic cells also have large numbers of protein channels and organelles specialised for internal transport.

Comparing prokaryotic and eukaryotic cells

There are a number of similarities and differences between prokaryotic and eukaryotic cells (Table 2.1.3). For example, both prokaryotes and eukaryotes contain DNA as their genetic material and use ribosomes to build their proteins. The shared features of prokaryotes and eukaryotes are evidence that supports the theory that they have a common ancestor. Figure 2.1.16 shows typical prokaryotic and eukaryotic cells and their features.

Prokaryotic cells are much smaller than eukaryotic cells, giving them a much larger surface area in comparison to their volume. This means that movement by diffusion into and out of prokaryotic cells is much faster than for eukaryotic cells. This results in prokaryotes having a higher metabolic rate, generally growing faster and having a shorter generational turnover. Bacteria can reproduce in between four and twenty minutes under ideal conditions.

TABLE 2.1.3 Comparison of prokaryotic and eukaryotic cells

Feature	Prokaryotic cells	Eukaryotic cells
Size	• very small	• larger, with large variation in size
Surface area-to-volume ratio (SA:V)	• large • allows materials to diffuse in and out of the cell rapidly	• smaller • results in slower diffusion
Membrane-bound organelles	• absent	• many organelles bound by membranes, forming an organised internal structure.
Chromosomal DNA	• DNA chromosome in the form of a single-stranded loop • located in a region of cytoplasm called the nucleoid, lacking a membrane	• DNA in the form of linear, thread-like chromosomes wound around proteins called histones. • located in the nucleus, which is separated from the cytoplasm by a double-layered membrane
Ribosomes	• many tiny ribosomes scattered in the cytoplasm.	• many ribosomes, either attached to the endoplasmic reticula, or free in the cytoplasm
Cell membrane	• bilayer of phospholipid molecules enclosing the cytoplasm in bacteria. • phospholipids are different and sometimes fuse into a monolayer in archaea. • contains fewer protein channels	• bilayer of phospholipid molecules enclosing the cytoplasm • contains many protein channels
Cell wall	• in bacteria, consists of a protein–carbohydrate compound called murein.	• present in fungi, plants and some protists • consists mainly of carbohydrates: chitin in fungi and cellulose in plants
Flagella	• may have flagella to provide movement • consist of three protein fibrils coiled in a helix and protruding through the cell membrane and wall	• may have flagella or cilia for motility (but not in fungi) • consist of a highly organised array of microtubules (hollow protein tubes) enclosed by the extended cell membrane

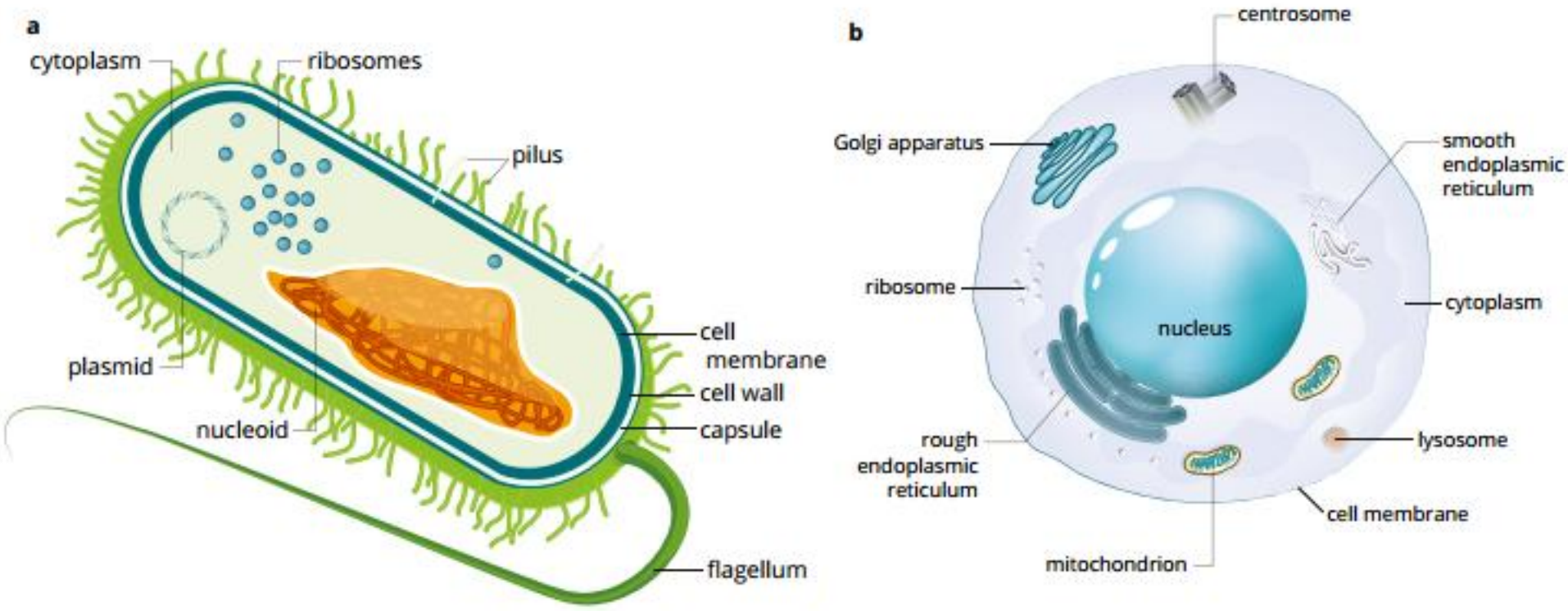


FIGURE 2.1.16 (a) A typical prokaryotic cell and (b) and a typical eukaryotic cell. Note the different membrane-bound organelles in the eukaryotic cell and the lack of such organelles in the prokaryotic cell. Prokaryotic and eukaryotic cells share some features, including a cell membrane, cytoplasm, DNA and ribosomes.

Both prokaryotes and eukaryotes have a cell membrane composed of a phospholipid bilayer with embedded proteins, which is further evidence of their common evolutionary relationship. However, eukaryotic membranes possess more protein channels than prokaryotes, which helps to increase the movement of materials across the membrane and offset the disadvantage that eukaryotes have when considering surface area-to-volume ratio.

Differences in the structures of the chromosomes means that eukaryotic cells have a much tighter control over gene expression than that seen in prokaryotes. This means that different genes can have different levels of activity, or none at all, in different cells. This differentiation in gene action results in cell specialisation and permits the development of multicellular organisms that have specialised tissues and organs. You will learn more about cell specialisation in Chapter 4.

Membrane-bound organelles allow eukaryotes to have a more complex metabolism than prokaryotes. This is because different processes can occur at the same time in different organelles, even if they require very different conditions to occur or would normally inhibit each other. The processes that occur inside organelles are also insulated from the external environment of the cell, so changes outside the cell are less likely to inhibit cellular processes.

Organelles that are involved in transport, such as Golgi apparatus and the endoplasmic reticula, allow eukaryotic cells to be much larger than prokaryotic cells as the use of transport channels makes movement of substances within the cell much faster. The presence of specialised channels means eukaryotes do not rely on diffusion alone for the movement of substances.

The attachment of ribosomes to a transport channel, the endoplasmic reticulum, results in a more efficient movement of proteins around the cell. The endoplasmic reticulum also provides isolated compartments in which particular environmental conditions can be maintained in order to form proteins into the shape needed for them to function correctly.

CELL REQUIREMENTS FOR LIFE

Like organisms, all cells have certain requirements. All life requires a source of energy. The amount of energy required depends on the type of cell, its stage of growth and its level of activity. Cells also require nutrients and water for growth, maintenance and repair, and constant environmental conditions to be maintained so that they survive and reproduce. Photosynthetic organisms can use inorganic materials from their environment to manufacture their own organic materials in the process of photosynthesis. All cells use these organic materials to produce energy in the process of cellular respiration. This relationship is shown in Figure 2.1.17.

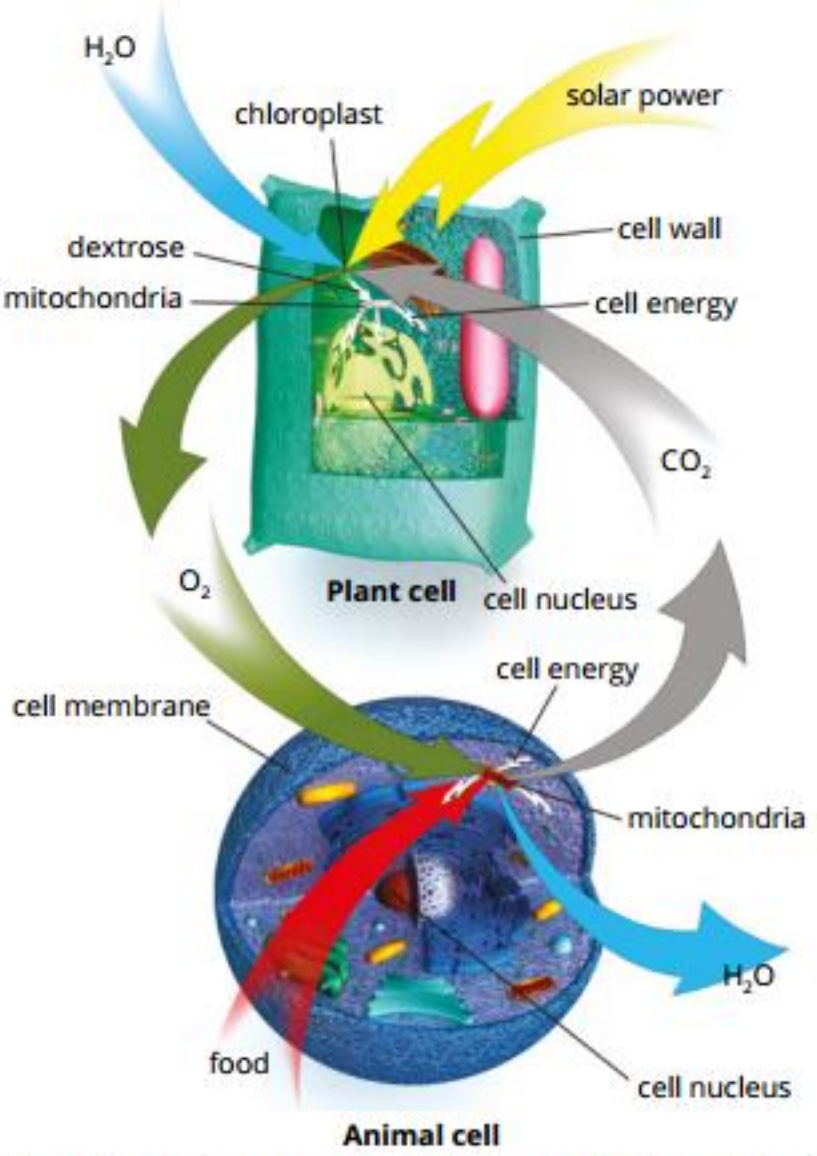


FIGURE 2.1.17 The relationship between photosynthesis and cellular respiration for the production of cellular energy in plant and animal cells. Plants take CO_2 from the environment and release O_2, while animals take O_2 from the environment and release CO_2.

The inputs and outputs of cells depend on the fundamental nature of the cell. Photosynthetic cells (some plant cells, protists and all cyanobacteria) and non-photosynthetic cells (all animal and fungal cells, some protists and bacteria) have different metabolisms. Photosynthetic cells make many of their requirements from simple inorganic substances, while non-photosynthetic cells must absorb complex chemicals for survival. Photosynthetic cells absorb carbon dioxide and water, along with minerals such as nitrates, and use these substances to make complex molecules such as carbohydrates, proteins, lipids and nucleic acids (DNA and **RNA (ribonucleic acid)**). Table 2.1.4 shows these compounds and their building blocks. Non-photosynthetic cells absorb the building blocks of these compounds, which they then use to make the compounds that they need.

TABLE 2.1.4 Essential compounds made by photosynthetic organisms and their building blocks

Compound	Building blocks	Made by photosynthetic organisms from
Carbohydrate (monosaccharides (Figure 2.1.18a), disaccharides (Figure 2.1.18b) and polysaccharides – Figure 2.1.18c)	simple sugars such as glucose, fructose and galactose (Figure 2.1.18b)	carbon dioxide and water
Protein	amino acids such as valine and leucine	carbohydrates and nitrogen from nitrates and other nitrogen-containing minerals
Lipid	fatty acids and glycerol	carbohydrates
Nucleic acids	nucleotides	carbohydrates and nitrogen from nitrogen-containing minerals

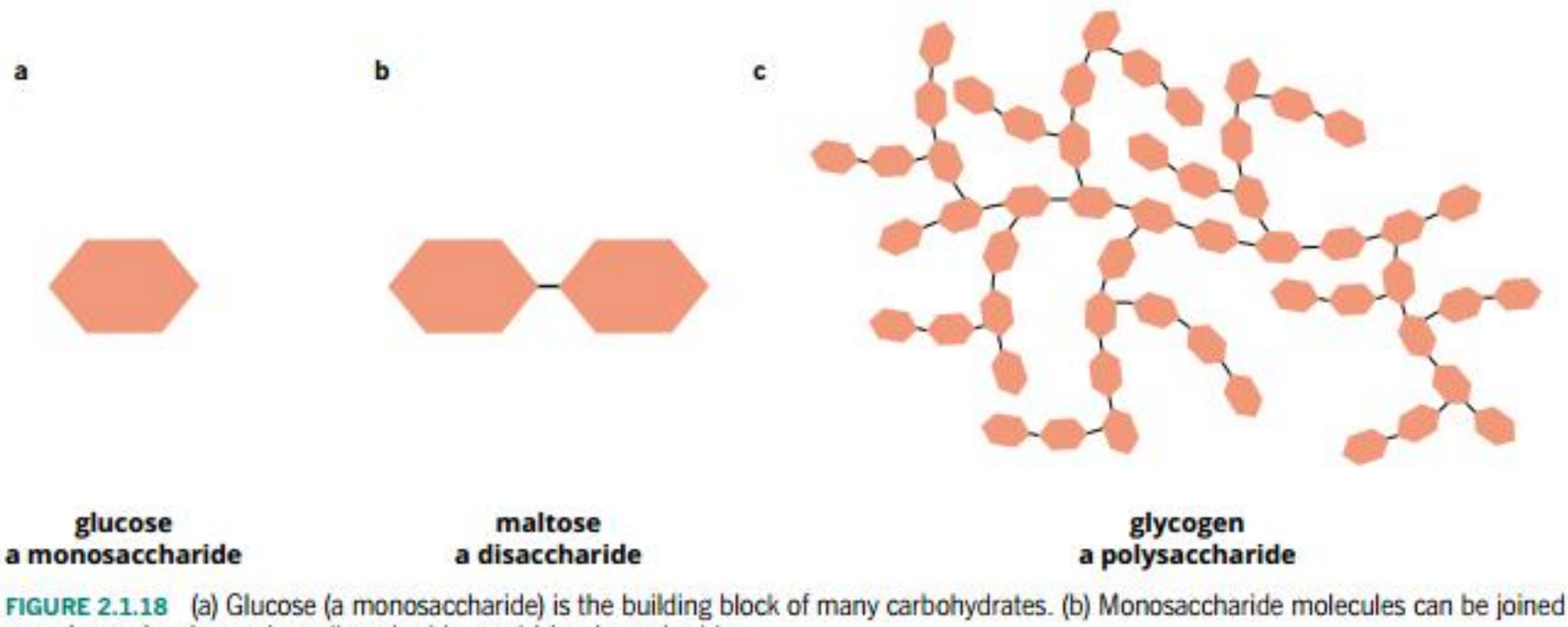


FIGURE 2.1.18 (a) Glucose (a monosaccharide) is the building block of many carbohydrates. (b) Monosaccharide molecules can be joined to make more complex molecules such as disaccharides and (c) polysaccharides.

Non-photosynthetic cells rely on the provision of more complex substances from other cells and organisms for their survival. Many cells can take these building blocks and modify them. For example, humans can make some **amino acids** by modifying other amino acids but are unable to make any from inorganic substances the way plants can. The amino acids are then used to build proteins.

In using energy (e.g. from monosaccharides, disaccharides and polysaccharides, lipids and proteins, all explained in Module 3.1) and carrying out the processes of growth, maintenance and repair, cells produce substances that are of no use to them or may be harmful to them. These waste substances (e.g. carbon dioxide, oxygen, urea, ammonia, uric acid, water and ions), along with metabolic heat, are often removed by releasing them into the external cellular environment. The ways that cells exchange substances with their environment depend on the type of material being exchanged. Cells must also be able to sense and respond to changes in their internal and external environments.

Proteins are large molecules composed of one or more polypeptides. Polypeptides are long, chain-like molecules consisting of many amino acids linked together.

2.1 Review

SUMMARY

- Living organisms have common characteristics and requirements—they are made of cells, are chemically complex and highly organised, exchange energy and materials with their environment, grow and reproduce, sense and respond to their environment, and show changes that are often adaptive.
- Cells use materials such as carbohydrates, proteins, lipids, vitamins and minerals to provide energy, and as raw materials for the formation of new structures, and produce wastes such as carbon dioxide, urea and excess heat.
- The cell theory is a fundamental principle of biology and is based on evidence collected over the last 300 years.
- The cell theory states that:
  - all organisms are composed of cells
  - all cells come from pre-existing cells
  - the cell is the smallest living organisational unit.
- All cells have a cell membrane, cytoplasm, genetic material in the form of DNA and ribosomes.
- There are two fundamentally different types of cells: prokaryotic and eukaryotic.
- Organisms with prokaryotic cells are called prokaryotes. They are classified into two domains: Bacteria and Archaea.
- Organisms with eukaryotic cells are called eukaryotes. They are classified into the domain Eukarya, which is divided into four kingdoms: Protista, Fungi, Plantae and Animalia.
- Prokaryotic cells have a simple structure, with a nucleoid lacking a membrane, scattered ribosomes, and DNA mainly in a single-stranded loop in the nucleoid.
- Eukaryotic cells have a complex structure, membrane-bound nucleus, many organelles in the cell cytoplasm, and DNA mainly in chromosomes in the nucleus.
- Archaea (the extremophiles) are often found in very harsh environments where their unique cell membrane structure protects them.
- Compartmentalisation (presence of membrane-bound organelles) in eukaryotic cells:
  - allows enzymes and reactants to be concentrated in particular organelles of the cell
  - maintains the right conditions for enzymes and reactants to function
  - allows incompatible chemical reactions to take place simultaneously within the cell
  - reduces the cell's vulnerability to environmental changes.
- Cells vary greatly in size, and a microscope is needed to see most cells.
- Laboratory research techniques include microscopy.
  - Light microscopes use visible light and a system of lenses to magnify images.
  - Electron microscopes use an electron beam focused by electromagnets to view objects. They have a much higher magnification and resolution than a light microscope.

KEY QUESTIONS

Describe

1 State the cell theory.

2 Name three components that all cells possess.

3 List three substances needed by cells to maintain life.

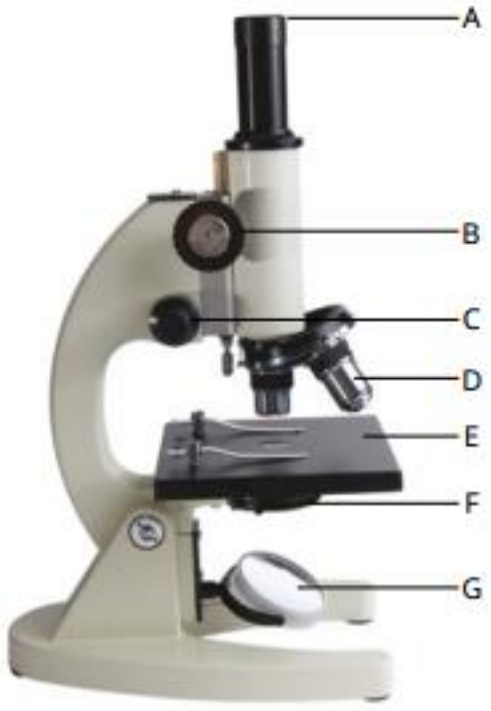
4 What are three wastes materials produced by cells?

5 Describe the differences between prokaryotes and eukaryotes.

6 Explain why eukaryotic cells can be significantly larger than prokaryotic cells.

7 Identify which kingdoms contain organisms that are composed of eukaryotic cells. Recall some examples from each kingdom.

8 Label the parts of the light microscope labelled A–G in the following diagram.



Apply

9 Draw and label a typical plant and animal cell.

10 Explain what is meant by 'cell specialisation'.

11 Explain what is meant by 'cell compartmentalisation'.

12 Outline the main differences between light microscopy and electron microscopy.

13 Convert 2.5 mm (millimetres) into µm (micrometres).

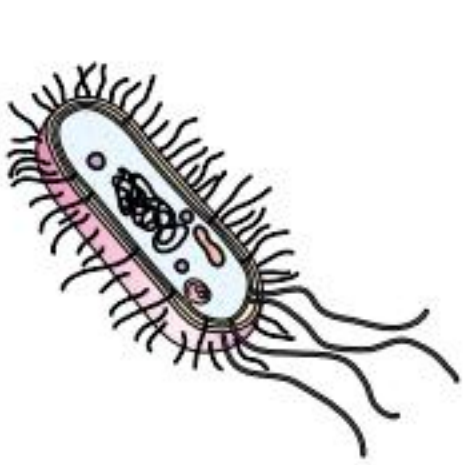
14 A structure is seen under a microscope. It is 0.5 µm long. Is this structure most likely to be a prokaryote cell, a eukaryote cell or an organelle from a eukaryote?

Analyse

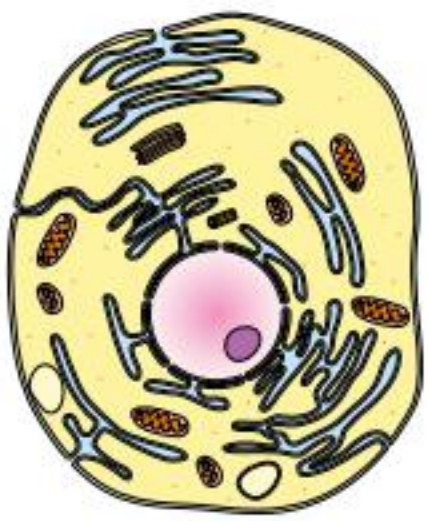
15 The following diagrams are of two cells observed with an electron microscope.

a Describe evidence that cell A is prokaryotic.

b Describe evidence that cell B is eukaryotic.



cell A (4500×)

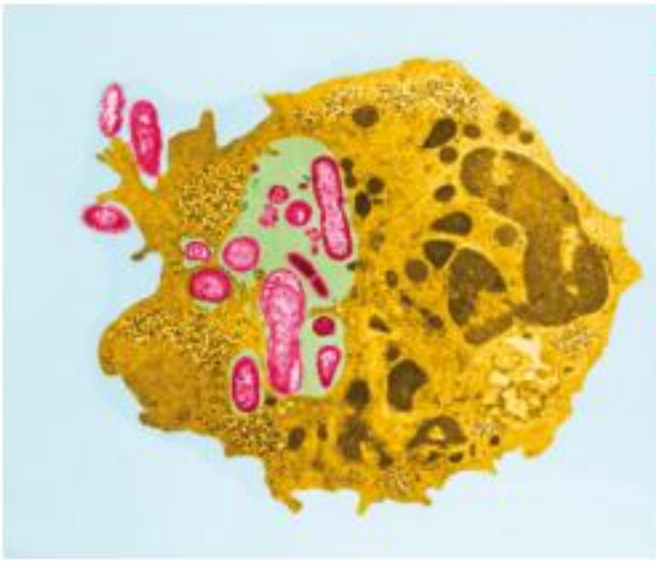


cell B (3000×)

16 Compare the structure of prokaryotes and eukaryotes.

17 Compare transmission electron microscopy and scanning electron microscopy.

2.2 Cell organelles



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- know and understand that eukaryotic cells have specialised organelles to facilitate cellular processes such as the synthesis of complex molecules, energy transformation, the storage of materials and the removal of cellular wastes and products
- identify key organelles and their functions, including the nucleus, mitochondria, rough endoplasmic reticulum, ribosomes, smooth endoplasmic reticulum, Golgi apparatus, lysosomes, vacuoles and chloroplasts
- identify organelles in electron micrographs.

You will recall from Module 2.1 that the two fundamentally different types of cells are prokaryotic and eukaryotic cells, and that organisms are classified into one of three domains (Bacteria, Archaea or Eukarya), according to their cell type.

Bacteria and archaea are prokaryotes. Their cells do not contain membrane-bound organelles. Prokaryotic cells have some non-membrane-bound organelles, such as ribosomes, a cell wall and sometimes flagella, although the structure and composition of these organelles are usually different from those of eukaryotic cells.

Animals, plants, fungi and protists are eukaryotes. Membrane-bound organelles are only present in eukaryotic cells. While eukaryotic cells may differ in appearance and function, they all contain membrane-bound organelles, such as those seen in Figure 2.2.1.

FUNCTION AND STRUCTURE OF ORGANELLES

Cells contain **organelles**, which have specialised functions. Organelles are subcellular structures involved in specific functions of the cell. Each organelle is functionally and structurally distinct. Most organelles are surrounded by membranes to separate their processes from other parts of the cell and to provide an optimal cellular environment for the biochemical reactions occurring in the cell.

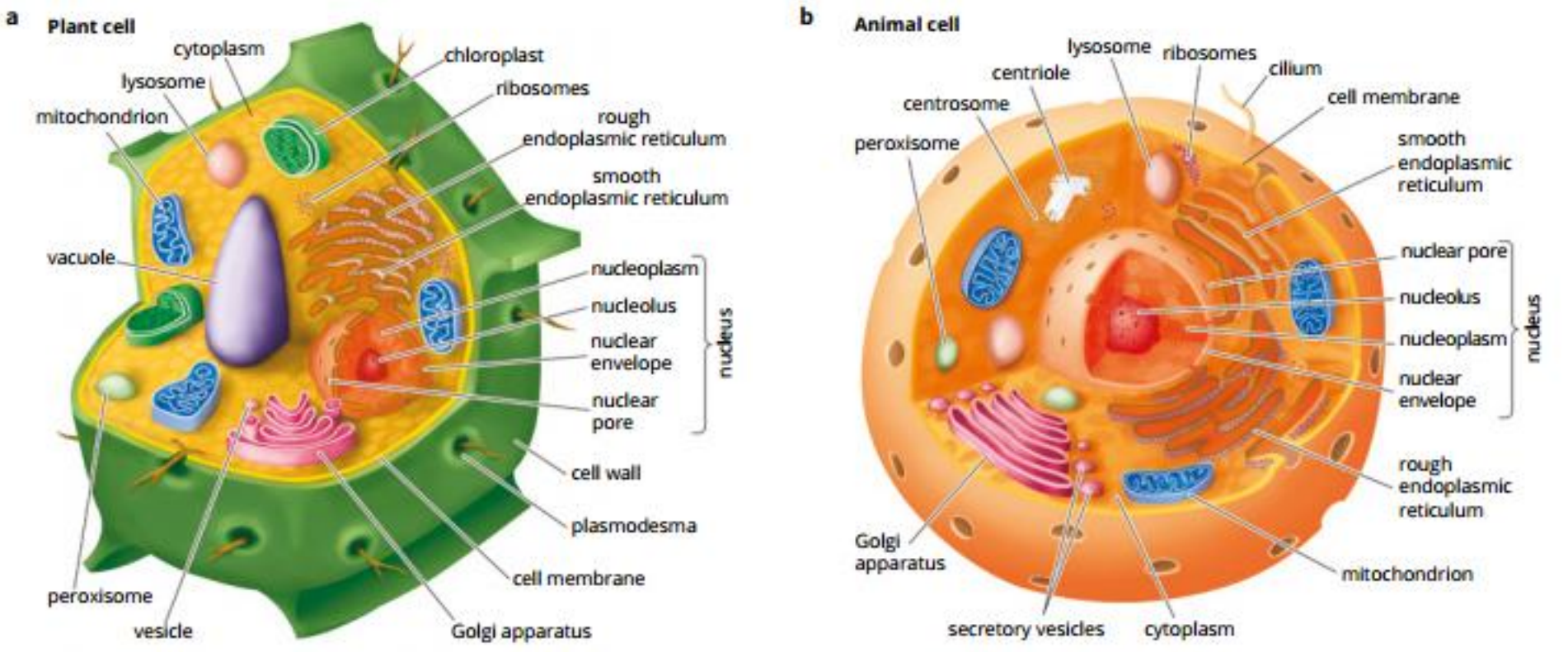


FIGURE 2.2.1 The many membrane-bound organelles of eukaryotic cells can be seen in these illustrations of (a) a plant cell and (b) an animal cell.

Prokaryotic cells do not contain any membrane-bound organelles. The number and types of organelles present in a cell depends on the function of the cell. Some organelles, such as chloroplasts, are only found in a limited number of types of cells, while others, such as ribosomes, are thought to be universal. Mitochondria, which liberate energy for cellular use, are found in very large numbers in active cells, such as muscle cells, and are relatively fewer in less active cells.

Typically, eukaryotes possess a nucleus, mitochondria, Golgi apparatus, rough and smooth endoplasmic reticulum, lysosomes and vacuoles. However, there are some exceptions, such as mature red blood cells, which lack both a nucleus and any lysosomes. Photosynthetic cells also contain chloroplasts (Figure 2.2.2). Eukaryotic cells also have many ribosomes. These may float freely in the **cytosol** or may be attached to the endoplasmic reticulum.

Each membrane-bound organelle has a different function, so each organelle requires a different internal composition, including a high concentration of enzymes and reactants.

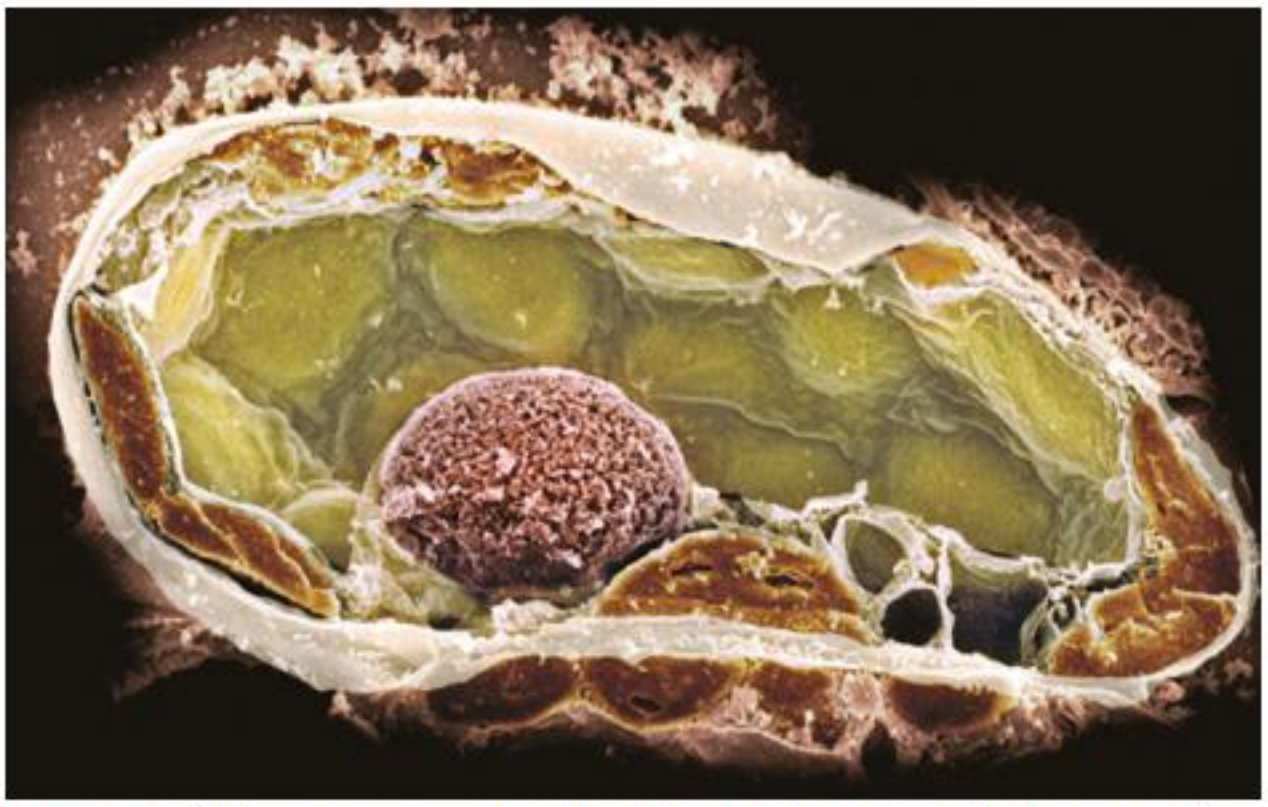


FIGURE 2.2.2 Plants are eukaryotes and so their cells have membrane-bound structures, some of which can be seen in this scanning electron micrograph. The cell wall is the external layer. Inside the cell wall are chloroplasts (brown), the nucleus (pink) and a large vacuole in the centre of the cell.

Enzymes are protein molecules that act as biological catalysts. Enzymes speed up the rate of reactions that would otherwise have taken place more slowly. Their action is specific: they catalyse only one type of reaction. You will learn more about the role of enzymes in cellular processes in Chapter 3.

Organelle membranes

The membranes surrounding organelles control the movement of substances between the organelle and the cell's cytosol. Just as cell membranes enable the cytosol to have a different composition from the cell's surrounding environment, membrane-bound organelles can have a different composition from the surrounding cytosol and other organelles. Because the environment on either side of the membrane is regulated, different types of biochemical reactions can occur in each region. Therefore, the metabolic reactions performed within the cells can occur efficiently within regions of optimal environmental conditions. The role of membranes to regulate cell function will be explored in Modules 2.3 and 2.4.

Cell compartmentalisation

Membrane-bound organelles in eukaryotic cells create compartments where specialised functions are carried out. This compartmentalisation of the cell ensures that chemical processes can occur efficiently. Compartmentalisation benefits the cell in several ways:

- It allows enzymes and reactants, for a particular function, to be close together in high concentrations and under the right conditions, such as optimum pH levels, so that the processes within the organelles are very efficient.
- It allows processes that require different environments to occur at the same time, in the same cell.
- It makes the cell less vulnerable to changes in its external environment, because changes affect the cytosol more than the membrane-bound organelles such as mitochondria or chloroplasts.

Organelles are involved in a number of different functions (Table 2.2.1). Their functions include the synthesis and processing of proteins and lipids, energy transformations, storage and maintaining the structure of the cell.

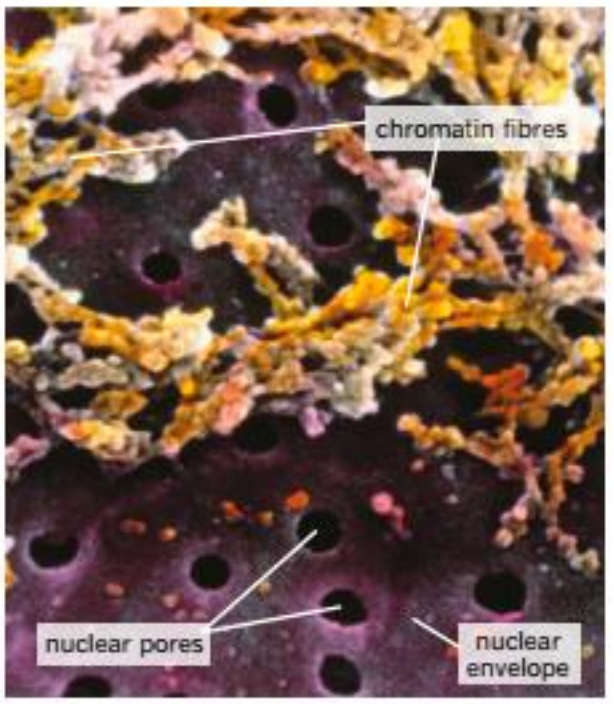


FIGURE 2.2.3 A scanning electron micrograph of the external surface of a nuclear envelope in an onion root tip cell. The envelope consists of a double membrane (purple), which encloses the nuclear DNA. The nuclear pores (black circles) are pathways for the transport of larger molecules between the nucleus and the cytoplasm. Contained within the nucleus are the chromatin fibres (yellow and orange), which contain the chromosomes.

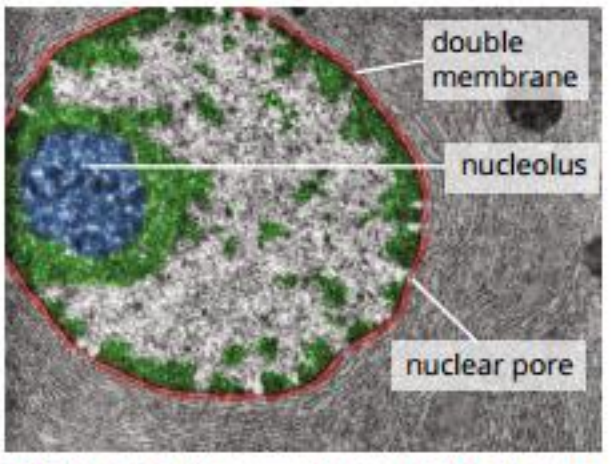


FIGURE 2.2.4 A coloured transmission electron micrograph of the nucleus of a plant. The double layered nuclear membrane (red), nuclear pores and nucleolus (blue) are clearly visible.

TABLE 2.2.1 Organelles and their functions

Function	Organelle	Present in plants	Present in animals
Involved in synthesis and processing of proteins and lipids	nucleus	✔	✔
	ribosome	✔	✔
	rough endoplasmic reticulum	✔	✔
	golgi apparatus	✔	✔
	lysosome	✘	✔
	smooth endoplasmic reticulum	✔	✔
Involved in energy transformations	mitochondrion	✔	✔
	chloroplast	✔	✘
Involved in storage and cell structure	centrioles	Sometimes	✔
	flagellum or cilium	✔	✔
	vacuole	✔	Small
	cell wall	✔	✘

Synthesis and processing of proteins and lipids

The following organelles are involved in the synthesis and processing of proteins and lipids in eukaryotic cells.

Nucleus

The **nucleus** of a eukaryotic cell contains the DNA, which carries the information needed for all cellular functioning. The genetic material in the nucleus takes the form of linear chromosomes composed of DNA and proteins. Chromosomes are usually not clearly visible, except during cell division. The nucleus is a large organelle and is easily viewed under a light microscope, however, an electron microscope is needed to see the nuclear membrane. The nucleus is surrounded by a double membrane, which regulates the flow of materials between the nucleoplasm and the cytosol. The nuclear membrane has pores through which substances can pass in a regulated way. These pores are shown in Figures 2.2.3 and 2.2.4. The outer layer of nuclear membrane is continuous with the endoplasmic reticulum.

The most visible structure inside the nucleus of a non-dividing cell is the **nucleolus**. The nucleolus is composed of proteins, DNA and RNA, and is where ribosomes are assembled.

Ribosomes

Cells contain many thousands of ribosomes, which are only about 30 nm in diameter and are therefore only visible using an electron microscope. Ribosomes are composed of proteins and **ribosomal RNA (rRNA)** and are sites of protein synthesis. They consist of two subunits joined together, as shown in Figure 2.2.5.

The subunits in eukaryote ribosomes are similar in structure but different in size from those in prokaryote ribosomes. Ribosomes are either free in the cytoplasm or bound to rough endoplasmic reticulum.

Ribosomes translate **messenger RNA (mRNA)** into proteins. Proteins produced in free ribosomes will function in the cell's cytoplasm, while proteins synthesised in ribosomes bound to the rough endoplasmic reticulum are secreted out of the cell, packaged into organelles or inserted in cell membranes.

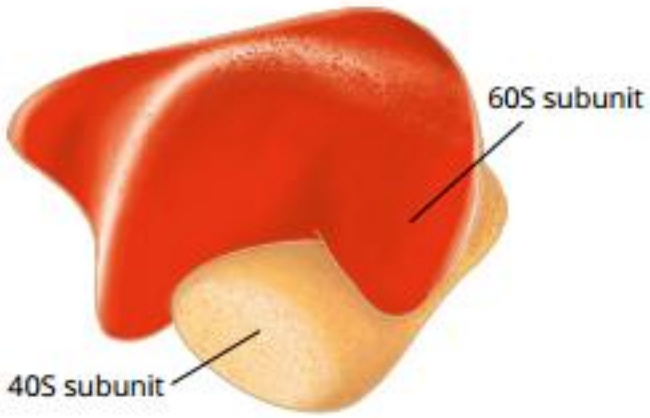


FIGURE 2.2.5 A single eukaryote ribosome consists of a larger 60S subunit and a smaller 40S subunit.

Endoplasmic reticulum

Endoplasmic reticulum is a network of intracellular membranous sacs (cisternae) and tubules that link with the cell membrane and other membranous organelles, including the nucleus. The endoplasmic reticulum can be rough or smooth.

Rough endoplasmic reticulum has ribosomes attached, which synthesise proteins. These ribosomes are bound to the membrane of the rough endoplasmic reticulum, as shown in Figure 2.2.6. After the proteins are made, they pass into the endoplasmic reticulum cavity containing enzymes. The enzymes add sugar molecules to the proteins to form glycoproteins.

Rough endoplasmic reticulum is abundant in cells that actively produce and export proteins, such as pancreatic cells, which secrete digestive enzymes. From the rough endoplasmic reticulum, proteins move into the Golgi apparatus for export from the cell.

Smooth endoplasmic reticulum contains the enzymes involved in the synthesis of molecules other than proteins, such as phospholipids and steroids. It is abundant in steroid-secreting cells in the testes, ovaries, kidneys and adrenal glands.

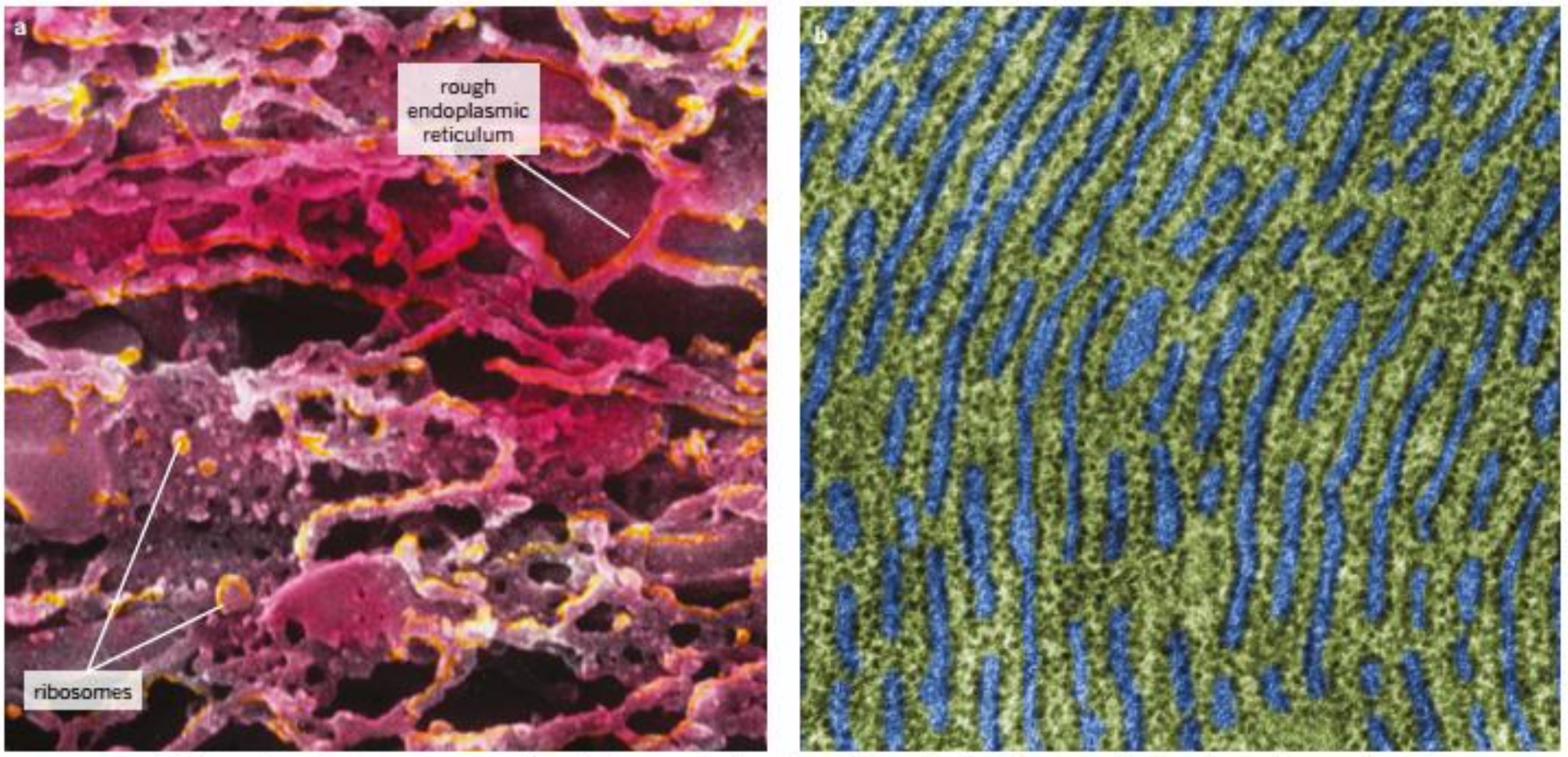


FIGURE 2.2.6 (a) A scanning electron micrograph of endoplasmic reticulum in a cell found in the olfactory epithelium (inside the nasal cavity). Endoplasmic reticulum is a network of folded membranes forming sheets, tubes or flattened sacs in the cell cytoplasm. On the surface of some of the endoplasmic reticulum membranes are ribosomes (yellow spheres). (b) Coloured transmission electron micrograph of rough endoplasmic reticulum (green) with ribosomes (faint black dots) and area inside the membrane, coloured blue.

Golgi apparatus

Figure 2.2.7 shows the **Golgi apparatus** (also called the Golgi body or Golgi complex), which is a stack of flattened smooth membrane sacs called cisternae. Unlike the rough endoplasmic reticulum, the cisternae in the Golgi apparatus are not connected. When proteins formed in the rough endoplasmic reticulum reach the Golgi apparatus, **vesicles** are formed from each cisternae to transport the proteins from one cisternae to the next. The proteins are modified for use by the cell, or for transport out of the cell. The cisternae then form transport vesicles to move these materials into the cytosol or out of the cell, such as secreted hormones. Vesicles budding from the Golgi apparatus also carry membrane-bound proteins to the cell membrane and digestive enzymes into lysosomes.

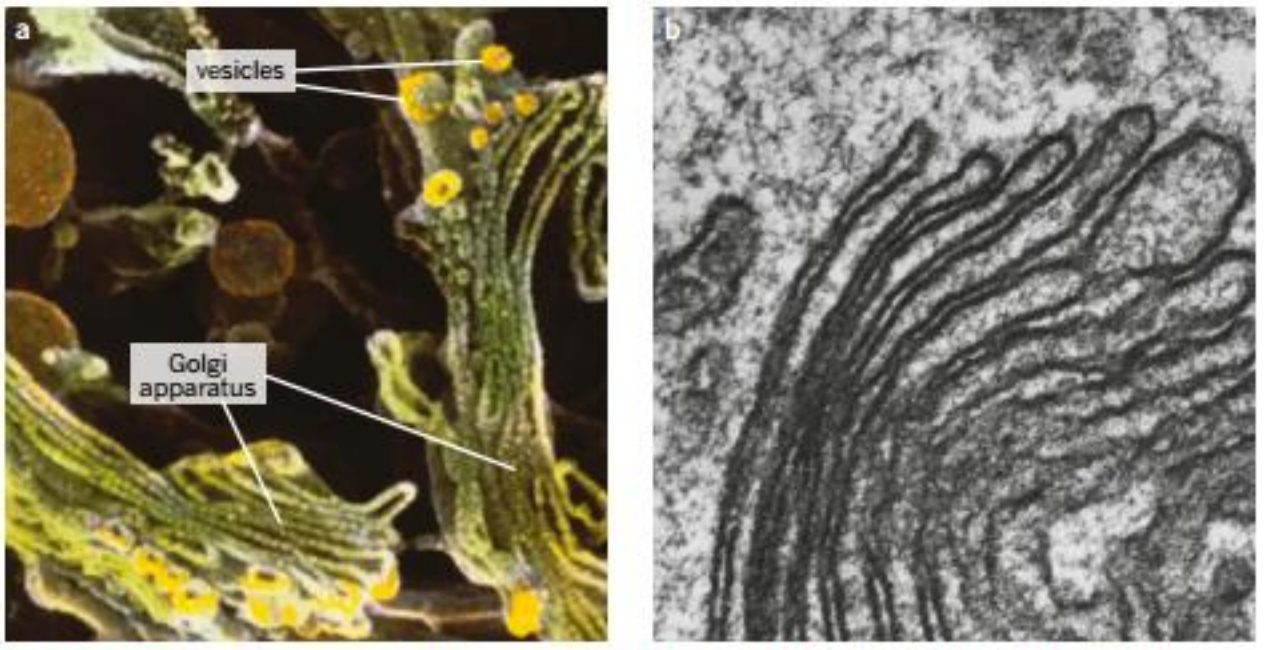


FIGURE 2.2.7 (a) A scanning electron micrograph of the Golgi apparatus of an olfactory bulb cell (part of the brain involved with smell). (b) A transmission electron micrograph of a Golgi apparatus. The Golgi apparatus consists of a stack of flattened interconnecting membranous sacs. It is the site in the cell of synthesis of biochemicals that are packaged into swellings at the margins of the sacs and become pinched off as vesicles (small yellow spheres in the (a)).

The Golgi apparatus has two faces: the *cis* face and the *trans* face, as shown in Figure 2.2.8. The cisternae of the *cis* face are connected to the endoplasmic reticulum, either directly or by small transport vesicles. This allows the proteins made in the rough endoplasmic reticulum to enter the Golgi apparatus.

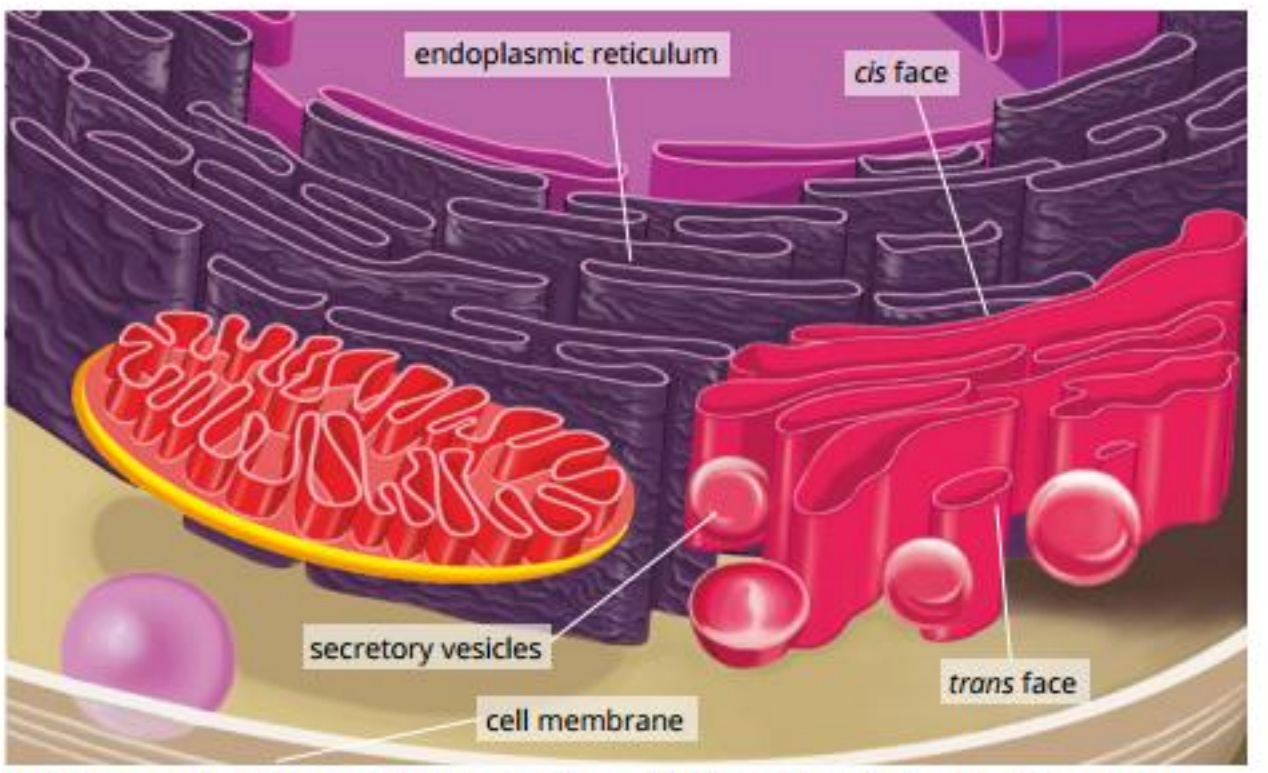


FIGURE 2.2.8 The Golgi apparatus has a *cis* face, which faces the endoplasmic reticulum, and a *trans* face, which faces the cell membrane.

The cisternae of the *trans* face are connected to the cell membrane by large secretory vesicles, which contain proteins to be secreted outside the cell. The membranes of the *cis* face more closely resemble the membranes of the endoplasmic reticulum, and the membranes of the *trans* face more closely resemble the cell membrane in their composition.

Secretory cells have a well-developed Golgi apparatus, but in other cells the Golgi apparatus is small. Some products packaged by the Golgi apparatus, such as the enzymes found in lysosomes, are not released from the cell.

Lysosomes

Figure 2.2.9 shows two **lysosomes**, which are specialised vesicles that digest (break down) unwanted matter. They are the recycling units of the cells. They are found only in animal cells. Lysosomes are formed when a transport vesicle containing enzymes is released from the Golgi apparatus and fuses with another vesicle called an endosome. The endosome contains molecules brought into the cell by endocytosis.

Lysosomes fuse with vesicles containing unwanted matter such as damaged organelles or foreign matter. The enzymes in the lysosome then digest the unwanted matter. Small molecules that the cell can reuse may diffuse back into the cytoplasm, but the rest are retained in the lysosome or released from the cell by exocytosis.

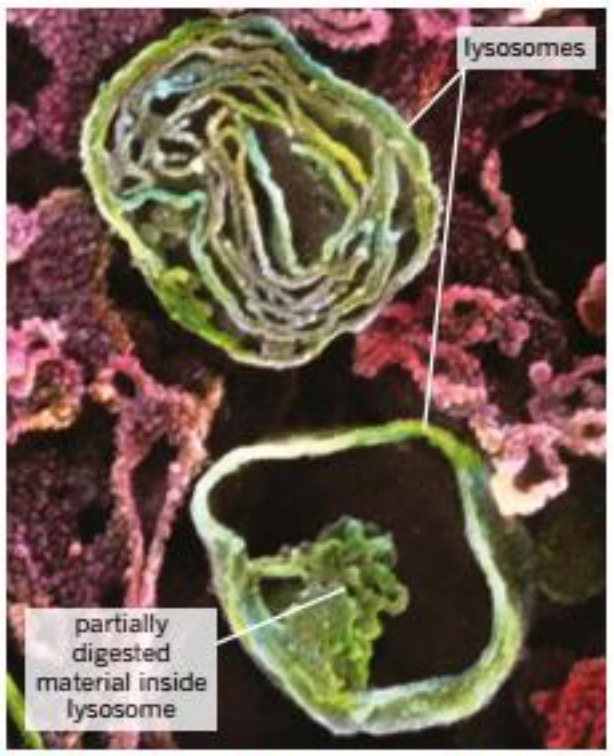


FIGURE 2.2.9 A scanning electron micrograph of two lysosomes in a pancreatic cell. Lysosomes (green) are small spherical vesicles bound by a single membrane (clearer on lower lysosome). Material that probably represents partially digested cell organelles can be seen in each lysosome.

Exocytosis is the fusion of a vesicle with the cell membrane, expelling its contents outside the cell.

Summary: synthesis and processing proteins and lipids

Protein and lipid synthesis and processing is shown in Figure 2.2.10. DNA is transcribed inside the nucleus into RNA. RNA moves out of the nucleus and binds to ribosomes. Ribosomes synthesise proteins using the information on the RNA. Proteins that are secreted out of the cell are made in the ribosomes bound to the rough endoplasmic reticulum. These proteins are modified and packaged in the Golgi apparatus. Vesicles arising from the Golgi apparatus fuse with the cell membrane, releasing their contents from the cell. They also insert membrane-bound proteins into the cell membrane. Lipids, such as glycolipids and steroids, are synthesised and processed in the smooth endoplasmic reticulum.

The presence of all of these transport structures makes movement of substances into, out of and around the cell efficient and relatively fast. It is this increase in efficiency that is one of the reasons that eukaryotic cells can be larger than prokaryotic cells.

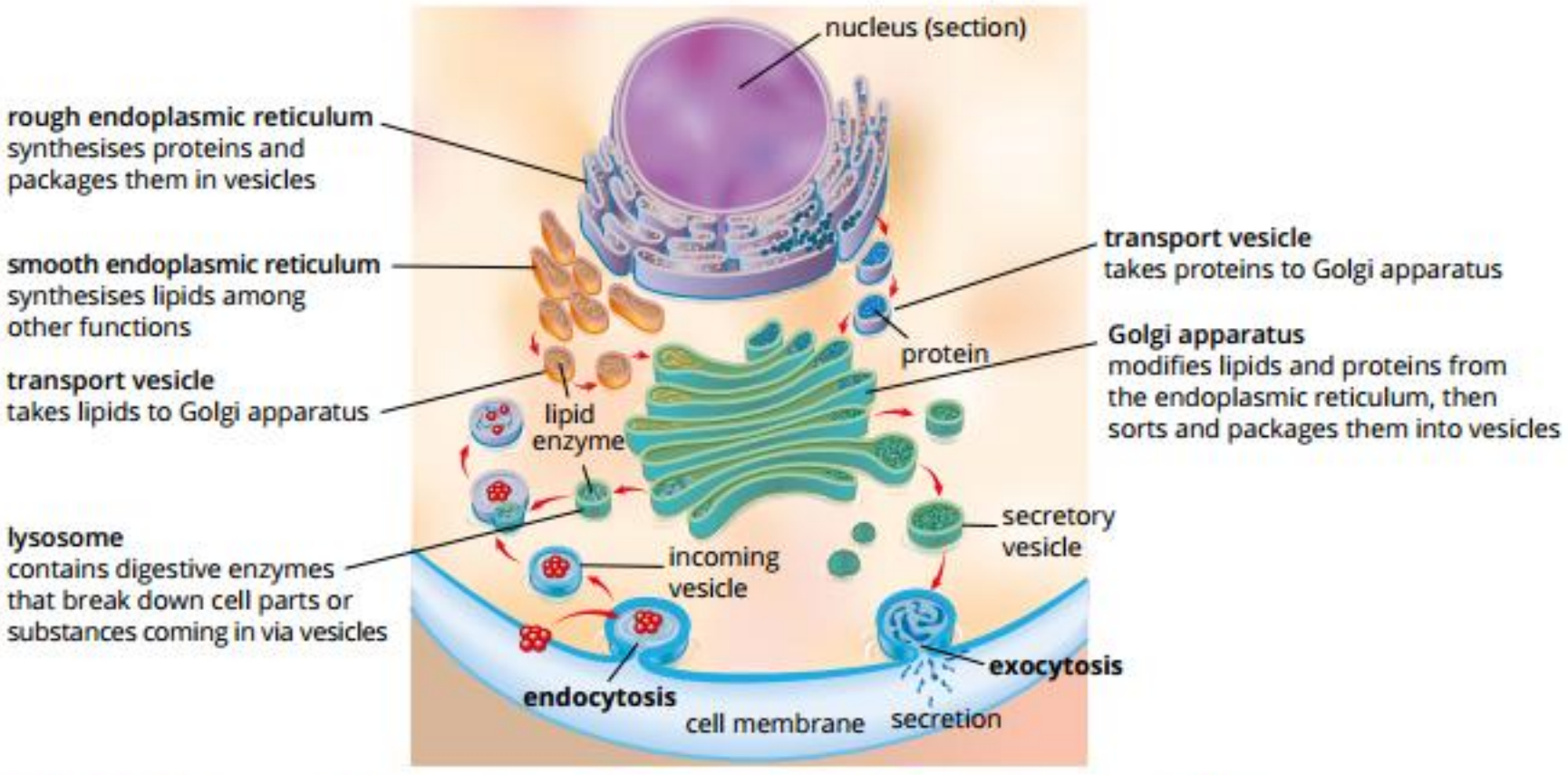


FIGURE 2.2.10 An animal cell, showing the organelles involved in synthesising and processing proteins and lipids

Energy transformations

Mitochondria and chloroplasts are the organelles involved in energy transformations within eukaryotic cells. Both mitochondria and chloroplasts contain their own DNA. They have a circular chromosome similar to that of prokaryotes. They also have their own ribosomes and make some of their own proteins.

Mitochondria

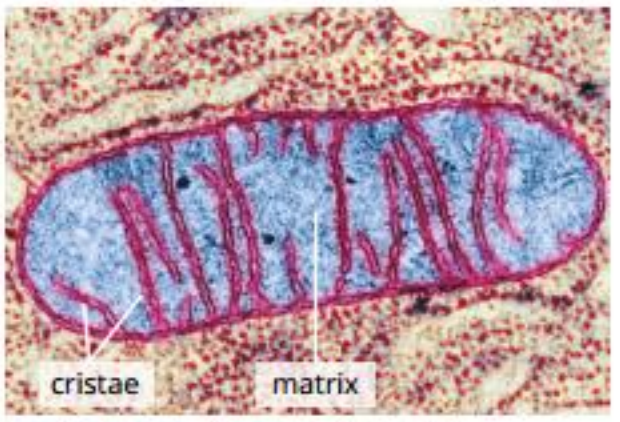


FIGURE 2.2.11 A transmission electron micrograph of a mitochondrion showing the matrix and cristae, which provide a large surface area for the reactions of aerobic respiration to occur.

Mitochondria are the main site of cellular respiration (Figure 2.2.11). In this organelle, complex organic molecules are broken down into simple molecules such as carbon dioxide and water. During this process energy is released. This energy is used for cellular processes such as cell division, making new organelles and building molecules such as proteins. The mitochondrion (singular) consists of two membranes. Both membranes regulate what goes into and out of the mitochondrion. The outer membrane completely surrounds the inner membrane and between the two membranes is the inter-membrane space. The inner membrane is relatively large and is very folded. The area inside the inner membrane is called the matrix. The matrix contains enzymes and other substances that allow the second stage of cellular respiration to occur (the first stage occurs in the cytosol). It is also where the mitochondrion's chromosome is found. The folds are called cristae. Embedded in the cristae are proteins that enable the final reactions of respiration to occur.

The mitochondrion is a fairly large organelle and can just be seen using a powerful light microscope, but an electron microscope is needed to see any of its internal detail.

Mitochondria are involved in the energy transformations that release energy from organic molecules for use by the cell. The number of mitochondria in a cell is related to the cell's energy requirements. Very active cells, such as heart muscle cells, have many thousands of mitochondria.

Chloroplasts

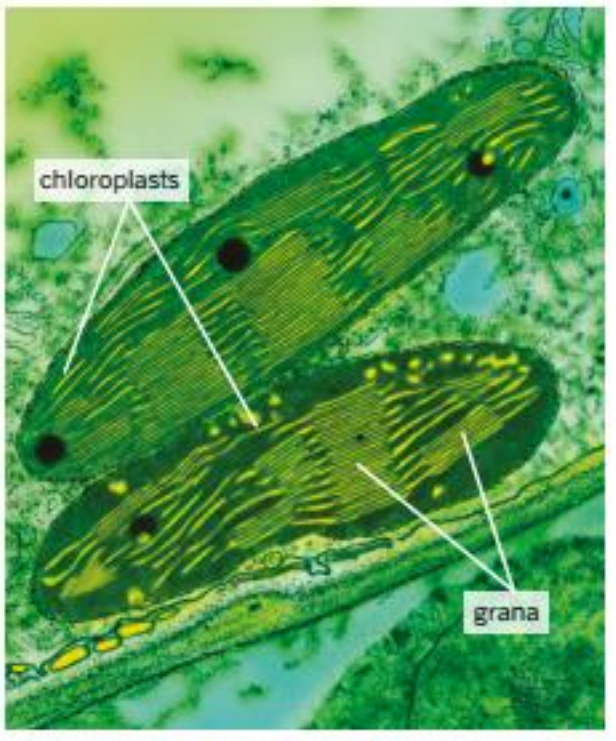


FIGURE 2.2.12 A transmission electron micrograph of two chloroplasts seen in the leaf of a pea plant, *Pisum sativum*. Each chloroplast is seen cut lengthways and contains stacks of flattened membranes (yellow) known as grana. The chloroplasts contain chlorophyll and are surrounded by an external double membrane.

Figure 2.2.12 shows **chloroplasts**, which are organelles involved with photosynthesis. They have a circular double-stranded DNA molecule and are green because of the large amounts of chlorophyll (a green pigment) they contain. They are present in plants and many protists, but never in animals or fungi.

Chloroplasts are composed of a system of three membranes: the outer membrane, the inner membrane and the thylakoid system. **Thylakoids** are disc-shaped sacs. This system of membranes forms compartments within the chloroplast that contain different enzymes. The stacks of thylakoids are the **grana** (singular granum), single thylakoids joining the grana are **lamellae** (singular lamella) and the fluid-filled spaces inside are the **stroma**. The thylakoids contain the light-trapping pigment, chlorophyll, and are where water is split into hydrogen and oxygen. In the stroma the hydrogen from the water and carbon dioxide from the air are used to build glucose. The oxygen is released into the atmosphere as a waste product.

Storage and cell structure

Vacuoles, plastids, cell walls, the cytoskeleton, centrioles, cilia and flagella are involved in storage and also support the cell structure in eukaryotic cells.

Vacuoles

Vacuoles are membrane-bound, liquid-filled spaces that store enzymes, and other organic and inorganic molecules. They occur in most cells, in different numbers. Figure 2.2.13 illustrates a plant cell vacuole. Vacuoles in animal cells and plant cells are different. Animal cells contain many small temporary vacuoles, but most plant cells contain a single large permanent vacuole surrounded by a membrane called the **tonoplast**. In plants, the vacuole provides structural support by helping maintain turgor and it seems that lysosome function also occurs here in the plant vacuole.

FIGURE 2.2.13 An illustration of a section through a plant cell, revealing its internal structure. At the centre of the cell is a large vacuole, which maintains the cell's shape, stores useful materials and digests the cell's waste products.

Plastids

Plastids are organelles involved in the synthesis and storage of different chemical compounds. They contain a double-stranded DNA molecule and possess a double membrane. Plastids develop from simple organelles called proplasts. Animal cells lack plastids. Plastids can be:

- chloroplasts, which are involved in photosynthesis and are found only in plants and some protists
- leucoplasts, which are involved in storage
- chromoplasts, which contain colour pigments and occur in petals and fruit.

Amyloplasts, as shown in Figure 2.2.14, are a type of leucoplast in plants. They are commonly responsible for synthesising and storing starch, but can also convert the starch back to sugar when the plant requires energy.

Tannosomes are a newly discovered organelle formed in chloroplasts and are the source of tannins. Tannins are polyphenols, and are part of the defence mechanism against bacteria and viruses in plants. Their bitter taste also discourages animals from eating the plants.

Cell wall

The cell wall is a rigid structure outside the cell membrane of plant cells, fungal cells and some prokaryotic cells. You can see the cell wall in a *Hookeria* moss cell in Figure 2.2.15. In plants, the cell wall is composed mainly of cellulose. The fungal cell wall is made of chitin.

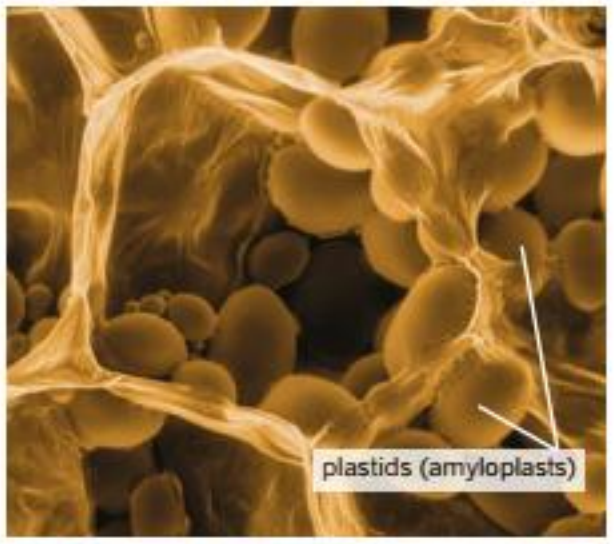


FIGURE 2.2.14 A scanning electron micrograph of amyloplasts (oval) in the sectioned cells of a potato (*Solanum tuberosum*). Amyloplasts are starch-storing plastids or plant organelles.

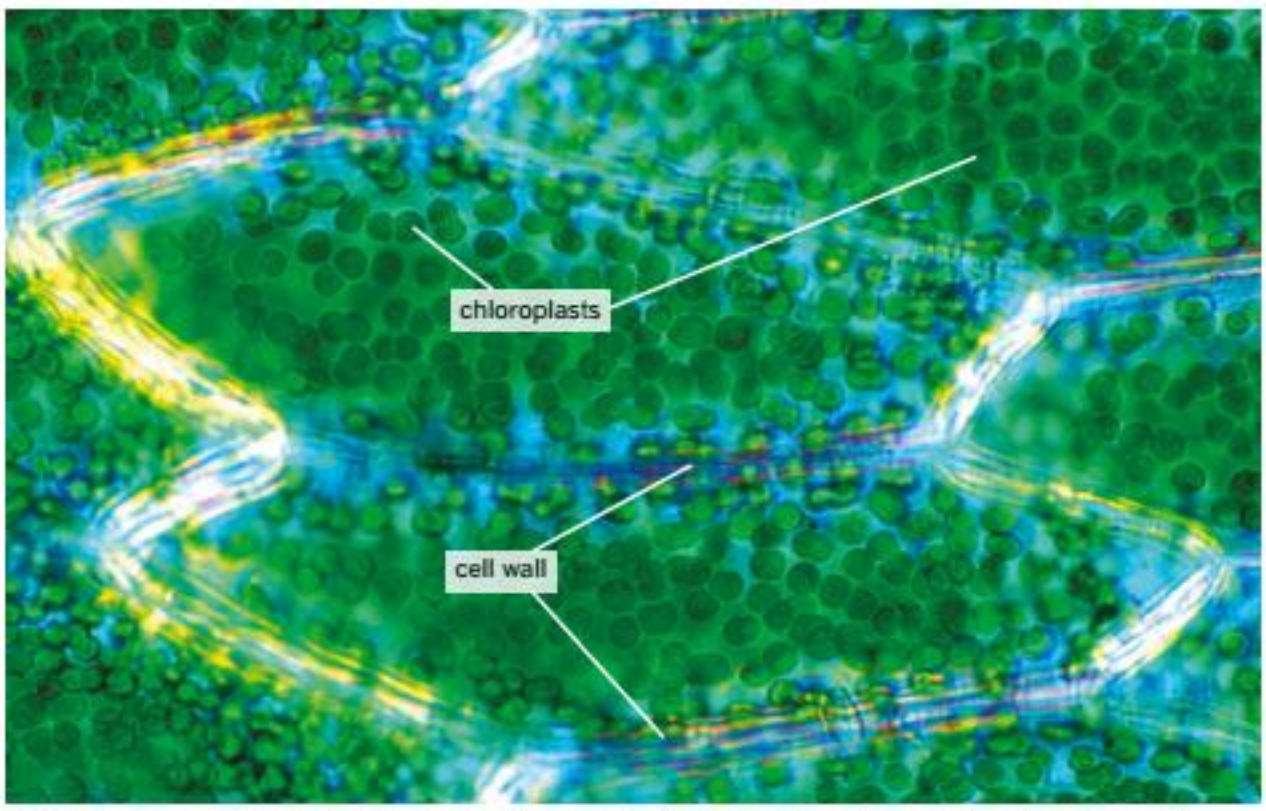


FIGURE 2.2.15 A light micrograph of cells in a leaf of shining *Hookeria* moss (*Hookeria lucens*). The leaf is made up of a single layer of cells. A cell wall (blue) encloses each cell, and numerous chloroplasts containing the pigment chlorophyll (green, round) are seen in each cell.

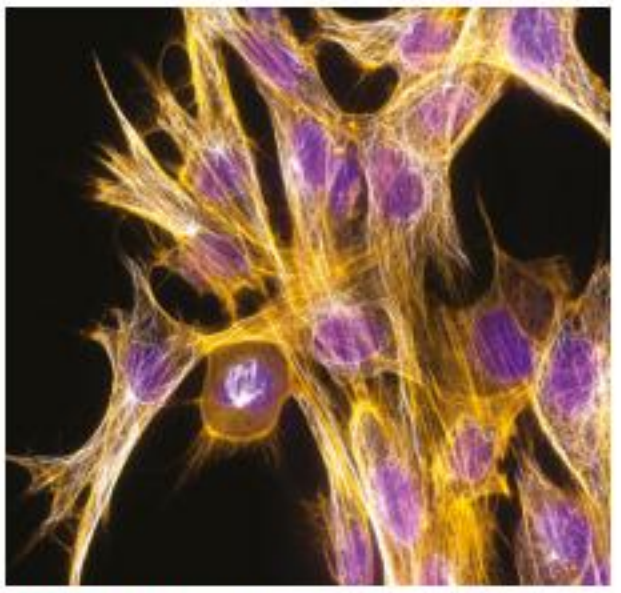


FIGURE 2.2.16 A fluorescent light micrograph of fibroblast cells. Fibroblasts are cells that give rise to connective tissue such as collagen, the main structural protein in the body. The proteins that make up the cytoskeleton are shown in yellow (actin) and white (tubulin). The cell nuclei are shown in purple.

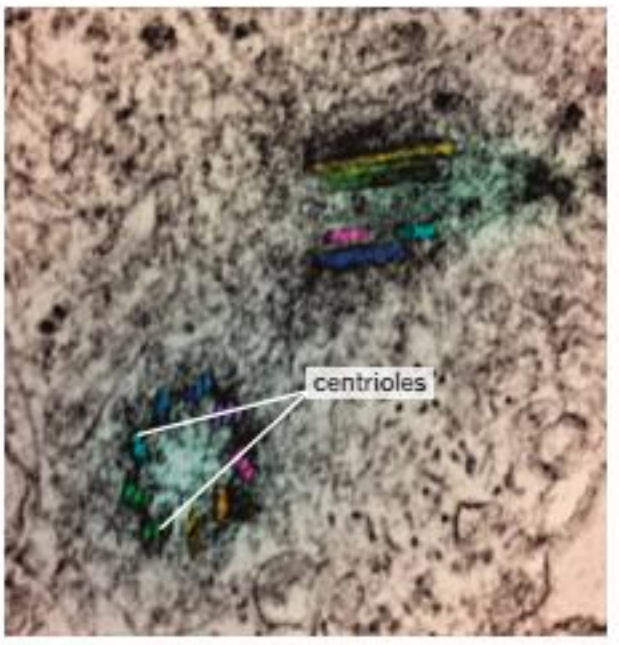


FIGURE 2.2.17 Transmission electron micrograph of centrioles (rainbow coloured structures), the organelles that are involved in cell division and the formation of some cell structures

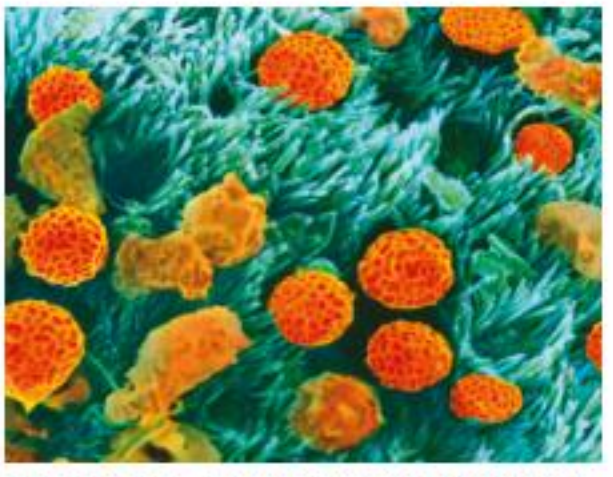


FIGURE 2.2.19 Scanning electron micrograph of the internal surface of the trachea (windpipe) with inhaled pollen (orange) and dust (brown). The surface cells have hair-like cilia (green) which, together with mucus, trap airborne particles for removal of foreign matter from the airways and lungs.

The cell wall provides support, prevents expansion of the cell and allows water and dissolved substances to pass freely through it. Lignin in the cell walls of woody plants gives them additional strength.

Cytoskeleton

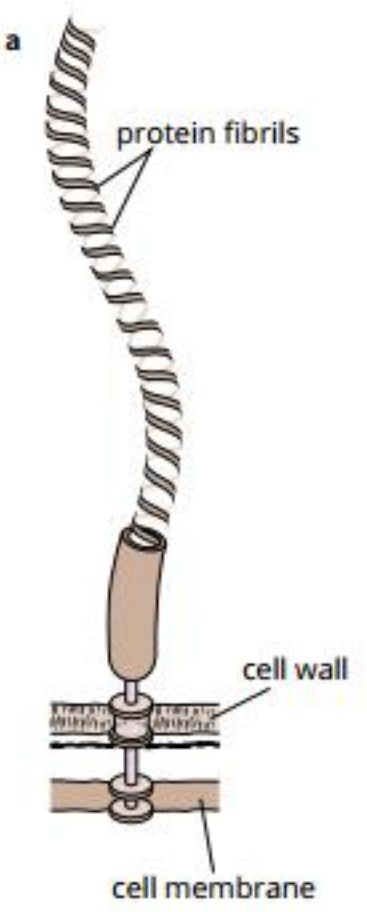
The **cytoskeleton** consists of microtubules of the protein tubulin and filaments of the protein actin. The cytoskeleton supports the cell's structure, allows the cell to move and assists in the transport of organelles and vesicles within the cell. Figure 2.2.16 shows fibroblast cells and their components.

Centrioles

Centrioles are a pair of small cylindrical structures composed of microtubules, as shown in Figure 2.2.17. They are present in most eukaryotic cells, but many plant cells do not have centrioles. Centrioles are involved in cell division and in the formation of cell structures such as cilia and flagella.

Cilia and flagella

Cilia and flagella (singular cilium and flagellum, respectively) are hair-like structures on the surface of cells and are shown in Figures 2.2.18 and 2.2.19. They consist of an arrangement of microtubules enclosed by an extension of the cell membrane. Cilia move with an oar-like motion and are usually shorter and more numerous than flagella. Both structures are involved in the movement of the cell.



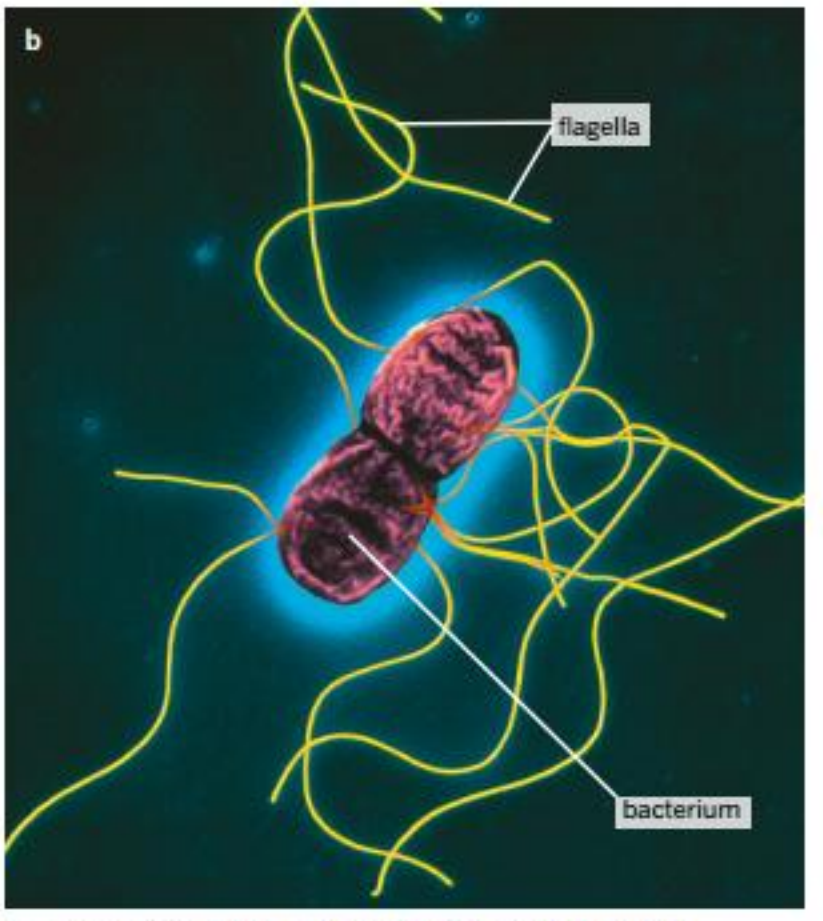


FIGURE 2.2.18 (a) Bacterial flagella consist of three protein fibrils coiled in a helical pattern. (b) A scanning electron micrograph of a *Salmonella typhimurium* bacterium. This rod-shaped, gram-negative bacterium moves by using its long, hair-like flagella (yellow).

A summary of the structure and function of the major cell organelles is given in Table 2.2.2.

TABLE 2.2.2 Summary of organelle structure and function

Organelle	Structure	Function
Nucleus	• membrane-bound: double membrane • contains DNA	contains hereditary information
Rough endoplasmic reticulum	• membrane-bound: network of cisternae • ribosomes bind to its membranes	processes and modifies proteins
Ribosome	• made of proteins and rRNA	synthesises proteins
Golgi apparatus	• membrane-bound: stack of cisternae that are not connected to each other	processes and packages proteins
Lysosome	• membrane-bound: vesicle containing digestive enzymes	digests cellular waste material and foreign matter
Smooth endoplasmic reticulum	• membrane-bound: network of cisternae	synthesises lipids
Mitochondrion	• membrane-bound: double membrane, inner membrane is highly folded • contains DNA	obtains energy from organic compounds
Chloroplast	• spherical or ellipsoidal, with double membrane • contains DNA and thylakoid sacs	uses light energy, carbon dioxide and water to produce glucose
Centriole	• small structure in the cytoplasm, consisting of microtubules	involved in cell division and the formation of cell structures such as flagella and cilia
Cilium or flagellum	• external structure consisting of microtubules	motility; movement of substances across cell surface
Vacuole	• membrane-bound, fluid-filled vesicle	stores substances; also involved in cell structure in plant cells
Plastid	• small, with double membrane • contains DNA	synthesises and stores various organic molecules
Cell wall	• external structure surrounding cell membrane • composition depends on type of cell	cell structure and protection

2.2 Review

SUMMARY

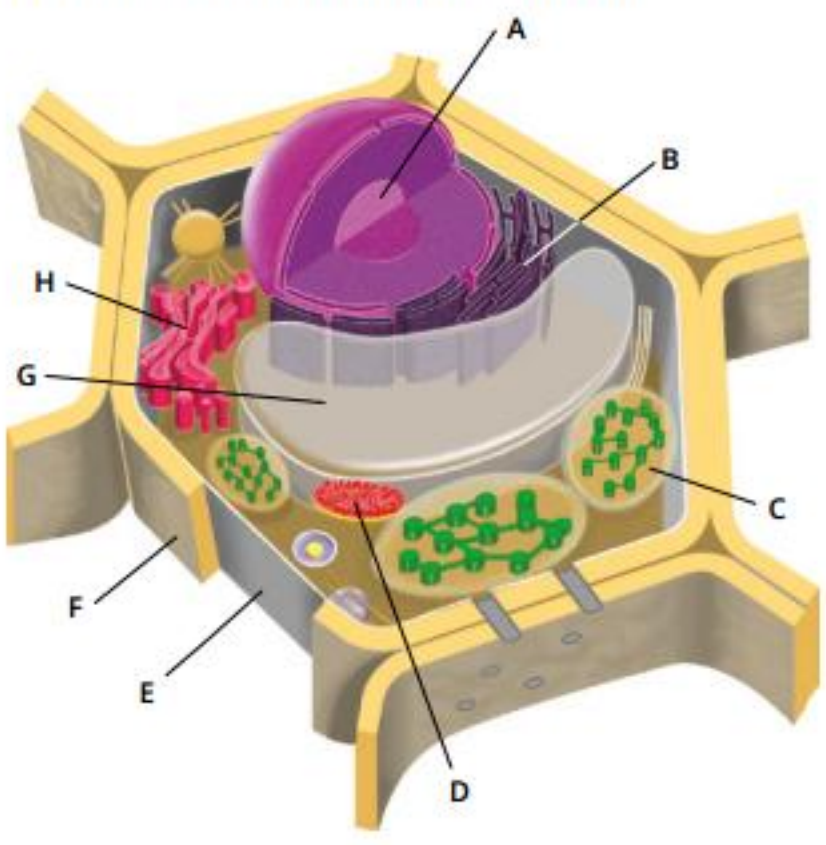
- Organelles are the functioning units of the eukaryotic cell.
- The main structures in a plant cell include the nucleus, tonoplast, vacuole, Golgi apparatus, rough and smooth endoplasmic reticulum, ribosomes, plastids, mitochondria and cell wall.
- The main structures in an animal cell include the nucleus, ribosomes, Golgi apparatus, rough and smooth endoplasmic reticulum, vacuoles, mitochondria, lysosomes, vesicles and centrioles.
  - The nucleus contains DNA and controls all cellular functions.
  - Golgi apparatus, rough and smooth endoplasmic reticulum, and vesicles are involved in transport of substances within the cell.
  - Ribosomes (proteins) and smooth endoplasmic reticulum (lipids) are responsible for synthesis of organic compounds for the cell.
  - Mitochondria and chloroplasts are responsible for energy transformations.
  - Lysosomes assist in the destruction of waste materials.

KEY QUESTIONS

Describe

1 What is the common role of mitochondria and chloroplasts in a cell?

2 Label the parts of the following plant cell.



3 Which organelles are responsible for each of the following functions?
- **a** protein synthesis
- **b** transport from Golgi apparatus to the cell membrane
- **c** lipid synthesis
- **d** storage
- **e** destruction of waste

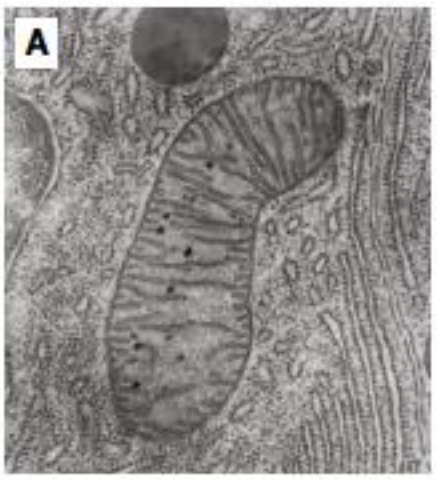
4 What function(s) do rough and smooth endoplasmic reticula have in common?

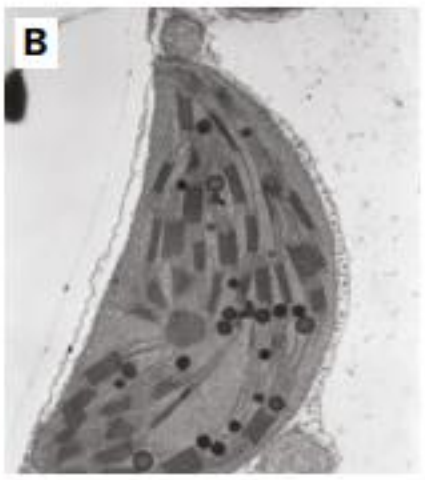
Apply

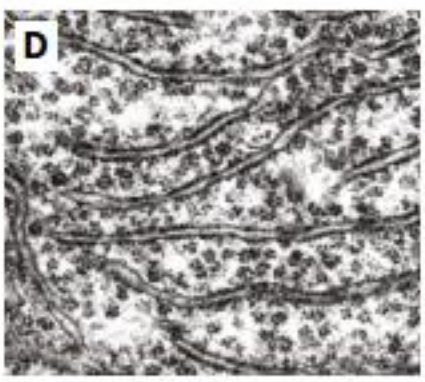
5 Which organelles would be most abundant in each of the following cell types? Explain your reasoning in each case.
- **a** enzyme-secreting cells
- **b** muscle cells
- **c** storage cells in a potato
- **d** cells that carry out photosynthesis in a leaf
- **e** cyanobacteria

6 Consider the following images. Identify which is a plant cell and which is an animal cell. Explain your reasoning.

7 For each of the images (A–D):
- **a** identify the name of the organelle
- **b** describe the organelle's function.







8 The cristae of the mitochondria and thylakoids of the chloroplasts provide large surface areas. Explain why this is advantageous for the cell.

9 The image below is of a mitochondrion as seen using a transmission electron microscope, but it is not how mitochondria typically appear. Explain why.

Analyse

10 You have been assigned the task of determining whether a sample contains plant or animal cells. Identify what features of the sample would help you in this task.

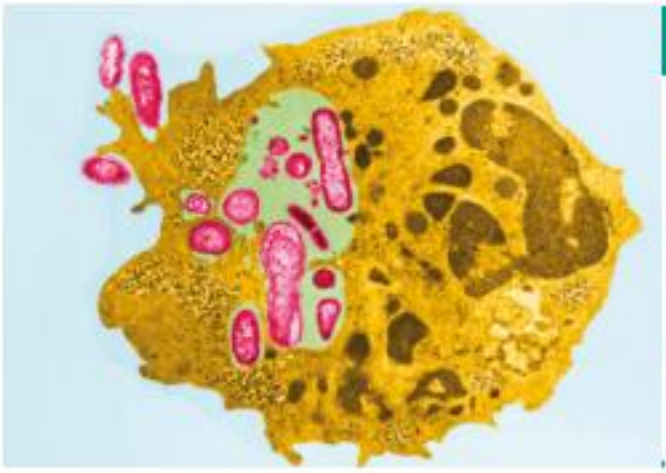
11 An experiment was conducted to investigate the synthesis of a type of organic substance from β-cells in the pancreas.

A radioactive material was injected into the secretory tissue. The level of radioactivity in various organelles of this type of cell was then measured every 60 minutes. The results are summarised in the table below.

	Percentage of total radioactivity			
Time (min)	**Rough endoplasmic reticulum**	**Golgi apparatus**	**Immature secretory vesicles**	**Mature secretory vesicles**
0	77	10	0	13
60	17	57	15	11
120	20	15	45	10
180	21	13	16	50
240	21	11	13	55
300	20	11	12	57

- **a** What type of substance is being synthesised?
- **b** What is the trend or pattern in radioactivity in the:
  - **i** endoplasmic reticulum
  - **ii** Golgi apparatus?

2.3 The cell membrane



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand the role of the cell membrane in the regulation of movement of materials into and out of the cell
- describe the structure of the cell membrane (including protein channels, phospholipids, cholesterol and glycoproteins) based on the fluid mosaic phospholipid bilayer model
- sketch a diagram of the phospholipid bilayer and explain the functions of each of the molecules that make up the membrane.

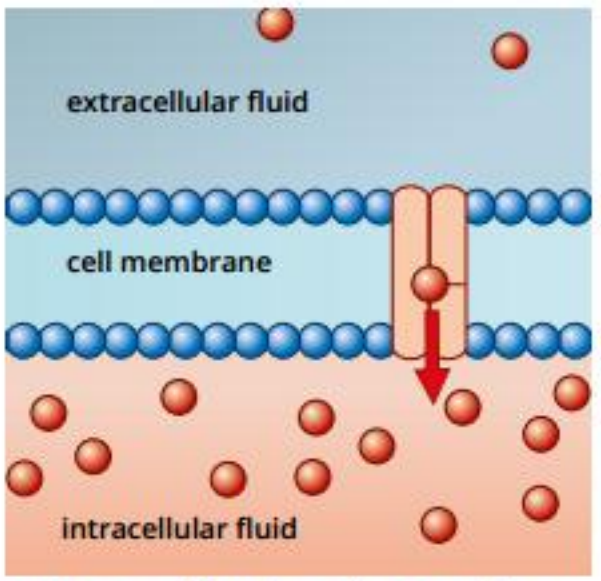


FIGURE 2.3.1 The cell membrane regulates the movement of substances between the extracellular fluid and intracellular fluid.

Extracellular fluid is fluid outside the cell membranes; it includes blood plasma and interstitial fluid.

FIGURE 2.3.2 Crabs have an external skeleton that protects them from water loss when on land.

In this module, you will learn about the composition and characteristics of the cell membrane. You will also study the ways cells can increase the surface area of the cell membrane available for the exchange of substances.

THE CELL ENVIRONMENT

Cells exist in a watery environment of **extracellular fluid**, which can be a large amount of fluid or a thin surface layer of fluid. In plants, the cell wall is porous and has little effect on the movement of molecules. All living cells exist in an environment that is a layer of fluid in contact with the outer cell membrane. The composition of this fluid is critical to the stability of cells because it is from this environment that cells obtain the nutrients they need. The cell membrane controls the movement of substances between the extracellular fluid outside and the **intracellular fluid** (or cytosol) inside the cell. A diagram of the cell membrane is shown in Figure 2.3.1.

Extracellular fluid in unicellular organisms

For unicellular organisms, the extracellular fluid is simply the watery external environment in which they live. Unicellular organisms can do little to control their environment and may die if it changes significantly.

However, some unicellular organisms such as yeasts can become dormant until their environment returns to optimal conditions. Other organisms can move slowly to a place where conditions are more suitable for their needs. For example, unicellular algae can move towards light, and some bacteria can detect and move towards nutrients or away from toxic substances.

Extracellular fluid in multicellular organisms

Conditions for cells in multicellular organisms are more stable than those of unicellular organisms. The more complex the organism, the more control it has over the environment in which its cells exist, and the more independent the organism is from its external environment. Whether they live in water or on land, multicellular organisms have an outer layer that acts as a protective barrier, such as a crab's exoskeleton as shown in Figure 2.3.2. This outer layer creates an internal environment for the organism that is different from their external environment, and organisms can better regulate their internal environment for optimal cell function. Therefore, in multicellular organisms, the environment of the cells is the extracellular fluid that surrounds them.

Most multicellular organisms can regulate their internal environment, often very precisely. This allows them to provide the specific conditions needed by specialised cells and tissues, and for their cells to function more efficiently. Commonly regulated aspects of the internal environment are:

- temperature
- oxygen concentration
- carbon dioxide concentration

- pH (acidity or alkalinity)
- osmotic pressure (concentrations of salts or ions)
- nitrogen waste concentration
- glucose concentration.

Importantly, the way cells interact with the extracellular fluid of the internal environment is regulated by the cell membrane.

CELL MEMBRANE COMPOSITION

Cell membranes have the same basic structure in all organisms, which serves to separate the interior of the cell (the cytoplasm) from its external environment. Most membranes are also asymmetrical, meaning one layer has different properties from the other. For example, the pattern of proteins and carbohydrate molecules in the external surface is different from the pattern in its internal surface.

The composition and characteristics of the cell membrane are related to the needs and function of the cell. The cell membrane performs important functions such as transporting molecules into and out of the cell, and recognising and communicating with other cells.

Phospholipids and the phospholipid bilayer

The cell membrane is a lipid–protein barrier that surrounds the cell and regulates the movement of materials between the inside and outside environment of a cell. It is typically about 7 nm wide. The **phospholipids** found in the cell membrane are composed of a phosphate group, a glycerol molecule and two **hydrophobic** fatty acid tails, as shown in Figure 2.3.3. The phosphate end of the molecule, which is polar and therefore **hydrophilic** ('water loving'), faces the aqueous regions of the cellular environment. The non-polar, fatty acid, part of the phospholipid is hydrophobic ('water hating') and faces inwards to the bilayer to form an oily region that becomes the barrier for most water-soluble materials.

A phospholipid is a molecule consisting of long-chain fatty acids (which are hydrophobic), a phosphate group and glycerol (which is hydrophilic). It is the major component of cell membranes.

Cell membranes are phospholipid bilayers that enclose the cytoplasm and subdivide the cell into compartments (organelles).

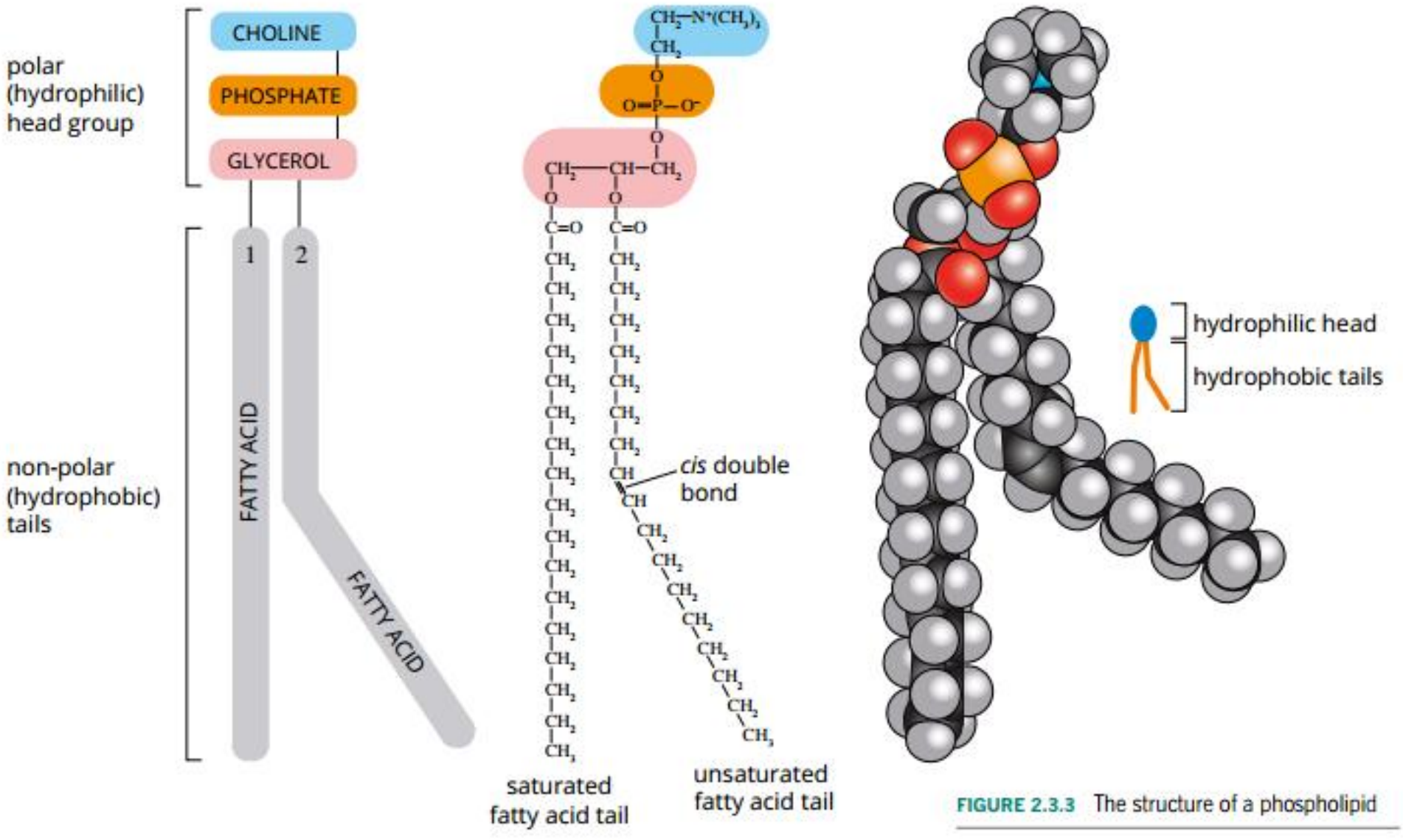


FIGURE 2.3.3 The structure of a phospholipid

The phospholipid bilayer of the cell membrane is called a bilayer because it has two layers of phospholipids. The hydrophilic heads form the outside and inside lining of the cell membrane, and the hydrophobic tails of the two layers of phospholipids meet in the middle. This is illustrated in Figure 2.3.4.

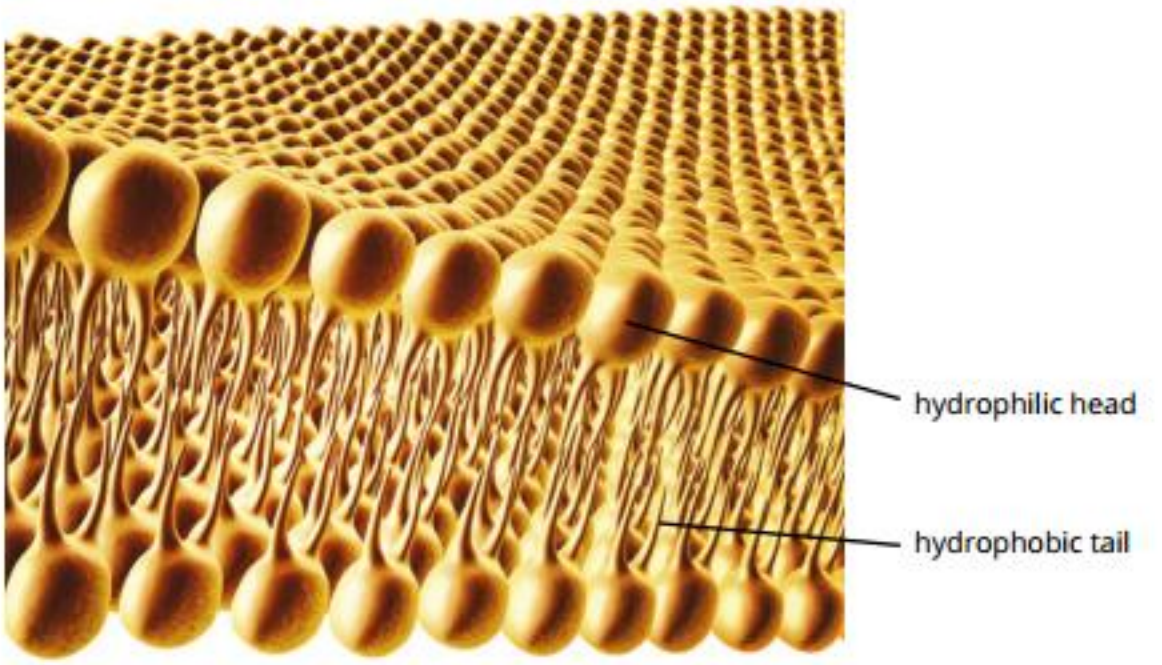


FIGURE 2.3.4 A phospholipid has a hydrophobic 'tail' and a hydrophilic 'head'.

Figure 2.3.5 shows how phospholipids arrange themselves in water and in oil. The phospholipids make cell membranes impermeable to water-soluble particles, ions and polar molecules. The movement of these molecules across the membrane is controlled by protein channels, which allow the cell to regulate the exchange of molecules with the environment. Controlling the movement of substances into and out of the cell is central to important processes that keep the cell alive, such as cell respiration, digestion and elimination of wastes. You will learn more about transport across cell membranes later in this module and in Module 2.4.

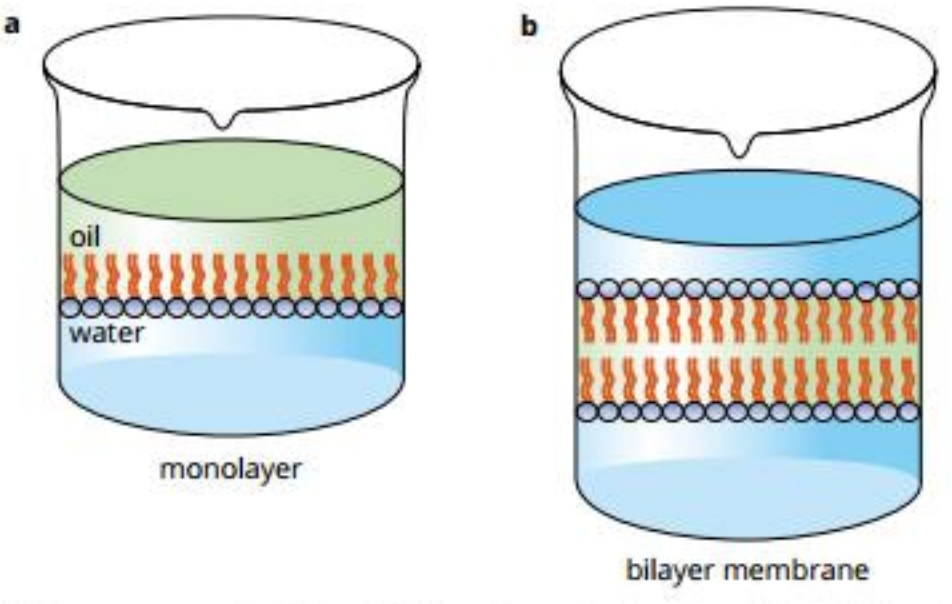


FIGURE 2.3.5 (a) The arrangement of phospholipids when water is on one side and oil is on the other side. (b) The arrangement of phospholipids when water is on both sides—a bilayer is formed.

Fluid mosaic model

Singer and Nicolson initially described the dynamic nature of the cell membrane as a 'fluid mosaic' in 1972. In their model, the phospholipids can move within the membrane by diffusion and a mosaic of discontinuous protein particles may be found floating in the phospholipid bilayer like icebergs. Figure 2.3.6 illustrates the fluid mosaic model.

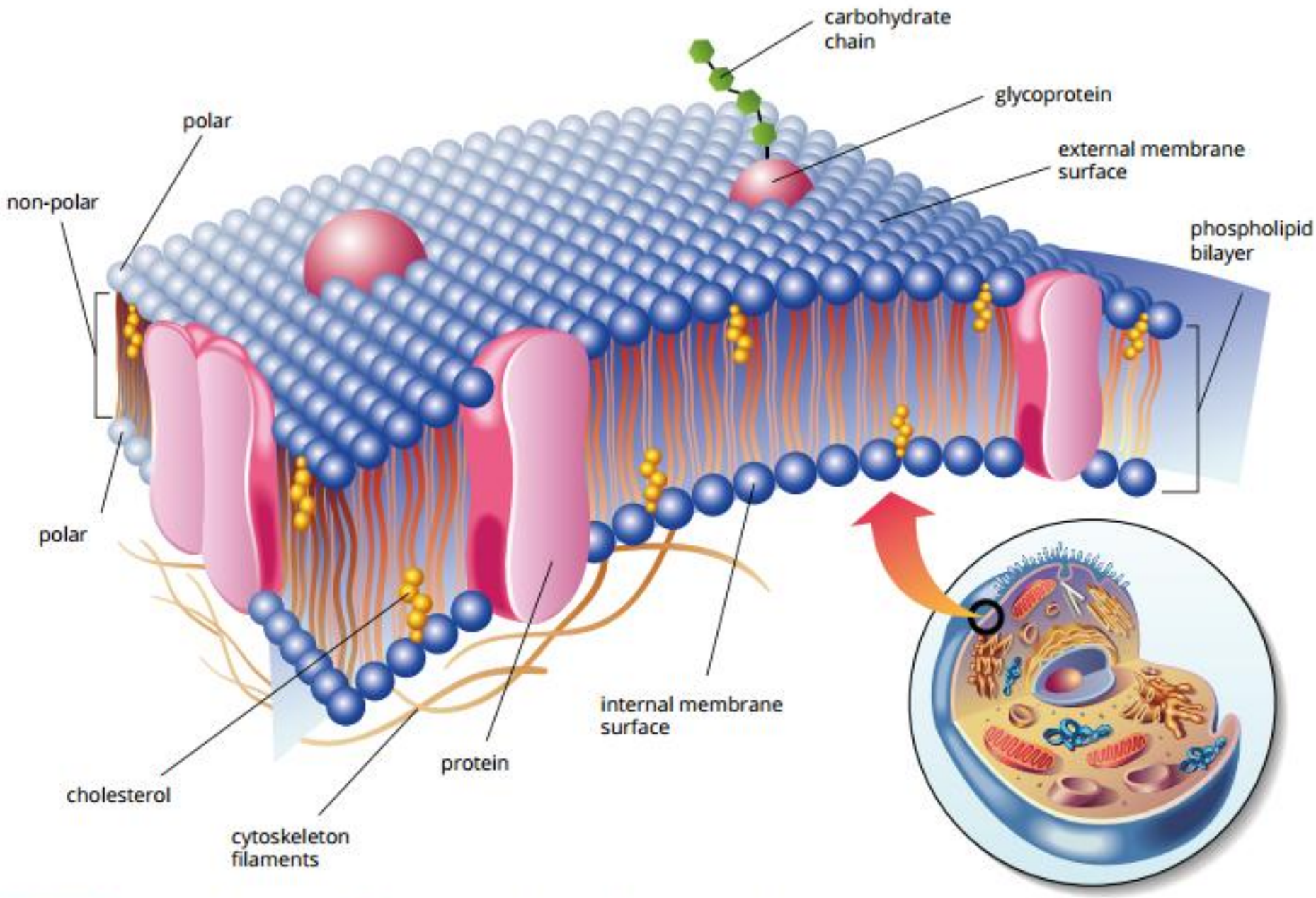


FIGURE 2.3.6 The fluid mosaic model of a cell membrane, showing the phospholipid bilayer in which large protein molecules are embedded

This fluid mosaic model is now widely accepted as the basic model of all biological membranes. According to this model, cell membranes consist of two layers of phospholipid molecules, with other molecules such as proteins, carbohydrates and cholesterol scattered throughout the membrane.

Molecules of the cell membrane are not fixed in place. Cell membranes are fluid structures, which means that individual phospholipid molecules (and some proteins) are free to move about within the layers. However, they rarely cross from one side of the membrane to the other. Most of the phospholipids and some of the proteins can move laterally, and sometimes some molecules are able to flip-flop transversely across the membrane. This movement is shown in Figure 2.3.7. The rate the molecules move within a layer of the cell membrane varies. Proteins in the membrane can move sideways throughout the membrane, but they move much slower than the phospholipids.

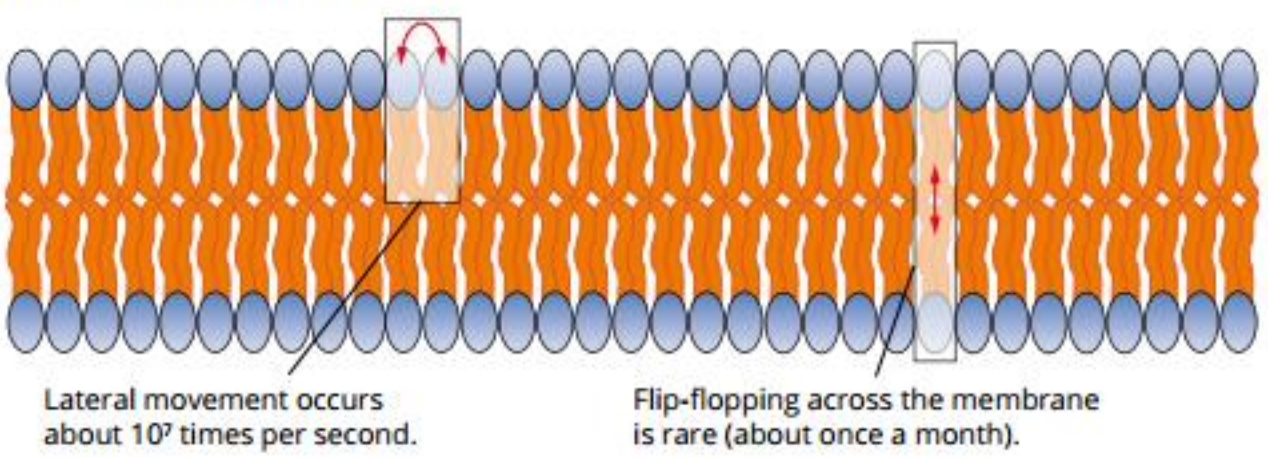


FIGURE 2.3.7 The movement of phospholipids in cell membranes

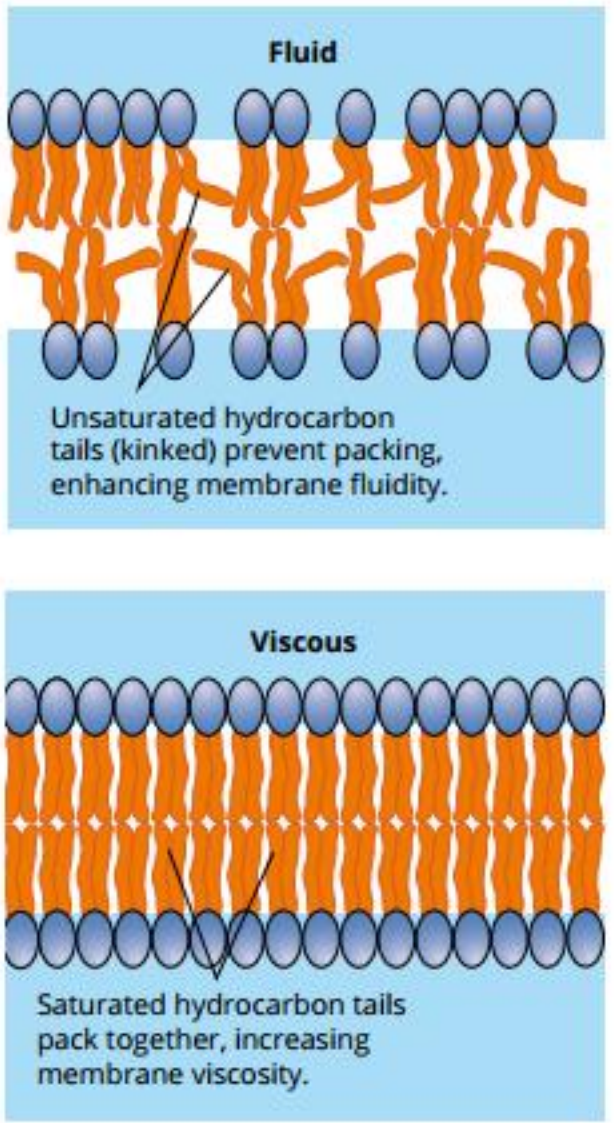

Fluid
Unsaturated hydrocarbon tails (kinked) prevent packing, enhancing membrane fluidity.
Viscous
Saturated hydrocarbon tails pack together, increasing membrane viscosity.

FIGURE 2.3.8 These two diagrams show the effects of unsaturated and saturated fatty acid tails on the fluidity of the cell membrane.

The ability of the phospholipids and proteins to move gives the cell membrane its fluid nature. Membrane fluidity is also influenced by how much unsaturated fatty acids are in the phospholipid molecules—more unsaturated fatty acids make the membrane more fluid, as shown in Figure 2.3.8. The fluidity of the cell membrane is very important because it affects how permeable the membrane is. It also makes it possible for proteins to move within the membrane to particular areas where they are required to carry out their function.

Figure 2.3.9 shows the components of the cell membrane. As well as the phospholipid bilayer, the cell membrane comprises cholesterol, proteins and carbohydrates.

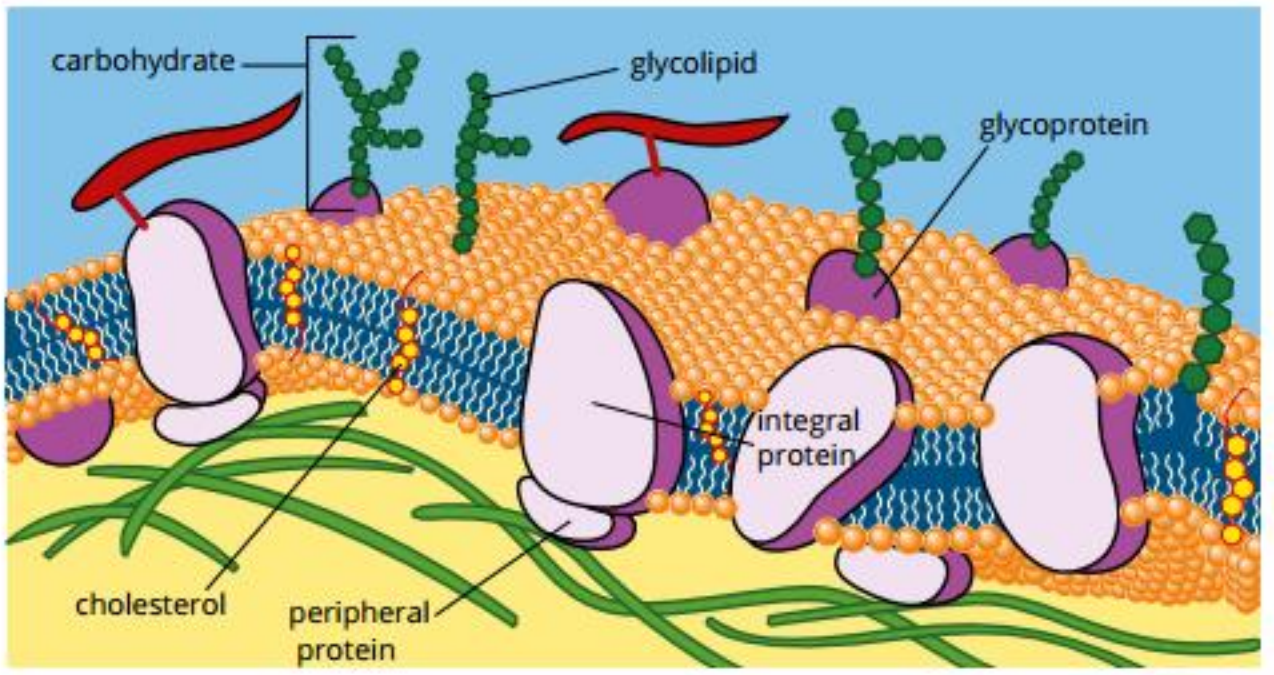


FIGURE 2.3.9 Components of the cell membrane

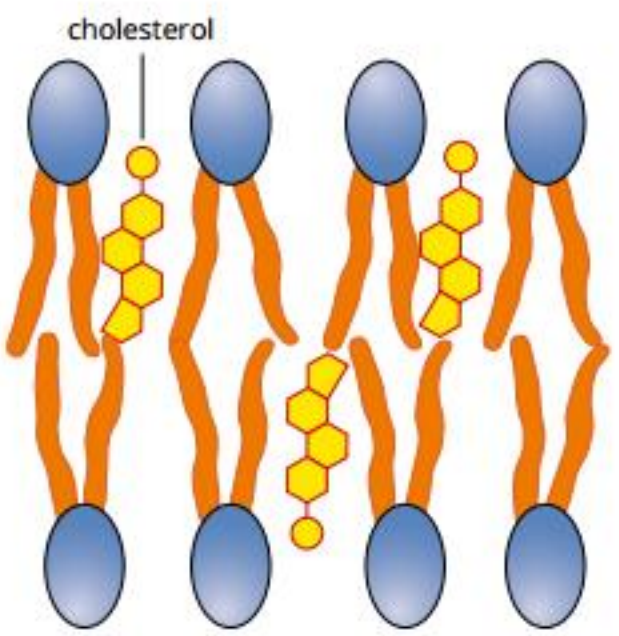


FIGURE 2.3.10 The cholesterol molecules are embedded within the hydrophobic fatty acid region of the cell membrane.

Cholesterol

Figure 2.3.10 shows how the cell membranes of eukaryotes contain **cholesterol**, a type of fatty molecule, between the phospholipid molecules. Cholesterol stabilises the membrane and reduces the permeability of the membrane to small water-soluble molecules.

Cholesterol acts as a buffer against changing temperatures. At high temperatures, cholesterol stops the cell membrane from becoming too fluid by restricting the movement of phospholipids. At low temperatures, cholesterol prevents the cell membrane from solidifying by restricting the tight packing of phospholipids.

Proteins

Like phospholipid molecules, proteins in the cell membrane can move about to some extent, but this movement may be limited to particular regions of the cell membrane.

Proteins that are a permanent part of the cell membrane are called **integral proteins**. Proteins that are a temporary part of the cell membrane are called **peripheral proteins**. Peripheral proteins bind to integral proteins or penetrate into one surface of the cell membrane (Figure 2.3.9). When integral proteins span both phospholipid layers, they are also called **transmembrane proteins**. Transmembrane proteins are involved in a number of important cellular and intercellular activities. These activities are illustrated in Figure 2.3.11.

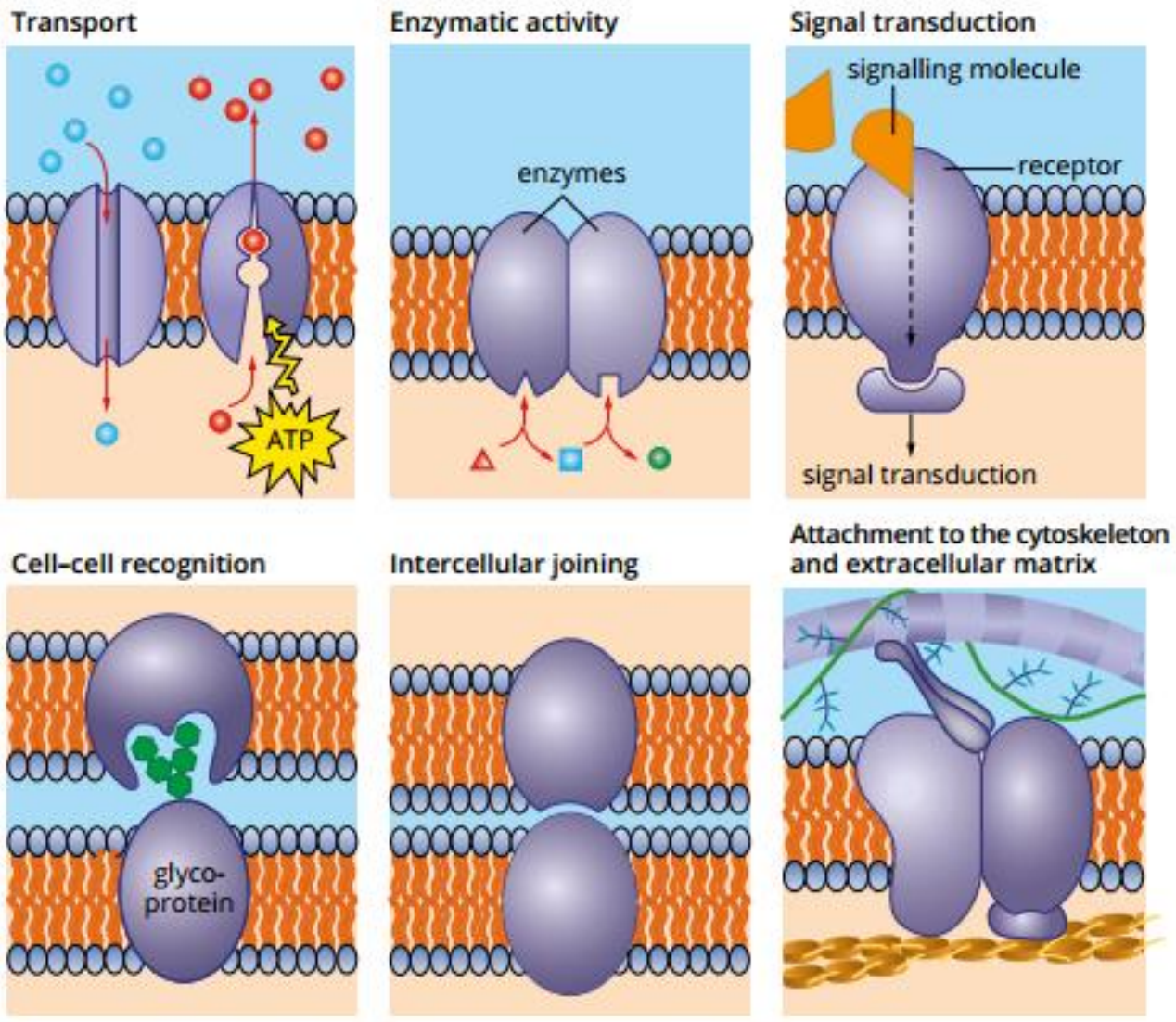


FIGURE 2.3.11 Different functions of cell membrane proteins

Integral proteins have many functions in the cell membrane (Figure 2.3.11). For example, these proteins:

- act as transport channels to transport molecules and ions through the membrane
- function as enzymes
- are involved in signal transduction (responding to hormones, neurotransmitters of other communication molecules from outside the cell)
- function in cell–cell recognition
- connect cells to each other
- act as attachments to the cytoskeleton and the extracellular matrix.

Protein channels

Protein channels are integral transmembrane proteins that enable the movement of polar and charged molecules to cross the cell membrane. Like all proteins, they are made of amino acids. Some amino acids are polar and are thus hydrophilic and lipophobic ('lipid hating'), and others are non-polar and are thus hydrophobic and lipophilic ('lipid loving'). The amino acids in a protein channel are arranged such that the outside of the channel has non-polar amino acids and the inside has polar amino acids. This creates an area inside the channel that is 'friendly' to molecules and ions repelled by the lipid bilayer of the cell membrane. The cell can regulate whether these channels are open or closed and can determine which substances can enter or leave the cell.

Carbohydrates

Carbohydrates associated with cell membranes are usually linked to protruding proteins (forming **glycoproteins**) or to lipids (forming **glycolipids**) on the outer surface of the membrane (Figure 2.3.9). They play a role in recognition and adhesion between cells, and in the recognition of antibodies, hormones and viruses by cells. Glycoproteins may act as receptors, receiving signalling molecules which trigger changes in cell functioning. They also often mark cells as self for recognition by white cells of the immune system.

2.3 Review

SUMMARY

- The external environment of living cells is the layer of extracellular fluid that is in direct contact with the cell membrane.
- For unicellular organisms, the extracellular fluid is the watery environment in which they live and that they can do little to control.
- Multicellular organisms have an internal environment that is more or less independent from the external environment. The external environment of the cells is therefore the extracellular fluid that surrounds them.
- Cell membranes separate the interior of the cell, the cytoplasm, from the external environment and control the movement of substances between the two.
- Cell membranes consist of a double layer of phospholipid molecules. They contain protein molecules of various sizes as well as fatty molecules such as cholesterol. They are also associated with other molecules, including carbohydrates.
- The phospholipid nature of the cell membrane makes it impermeable to water-soluble particles, ions and polar molecules.
- Cell membrane proteins:
  - provide selective channels that enable water-soluble particles and ions to travel through the cell membrane
  - catalyse reactions associated with the cell membrane
  - communicate with the external environment and other cells
  - bind with other cells.

KEY QUESTIONS

Describe

1 List three functions of the cell membrane.

2 What is the component of the phospholipid molecule that does not allow most water-soluble substances to pass through the phospholipid bilayer of the cell membrane?

3 How does cholesterol help maintain the cell membrane's structure?

4 What is an integral protein?

Apply

5 **a** The illustration below shows the structure of a typical cell membrane. Identify the compounds labelled A and B.

b With specific reference to the structure of compound A, describe how it is organised to form a bilayer.

6 Explain the importance of the fluidity of the cell membrane for the function of eukaryotic cells.

7 Explain the role the cell membrane plays in regulating the internal environment of a cell.

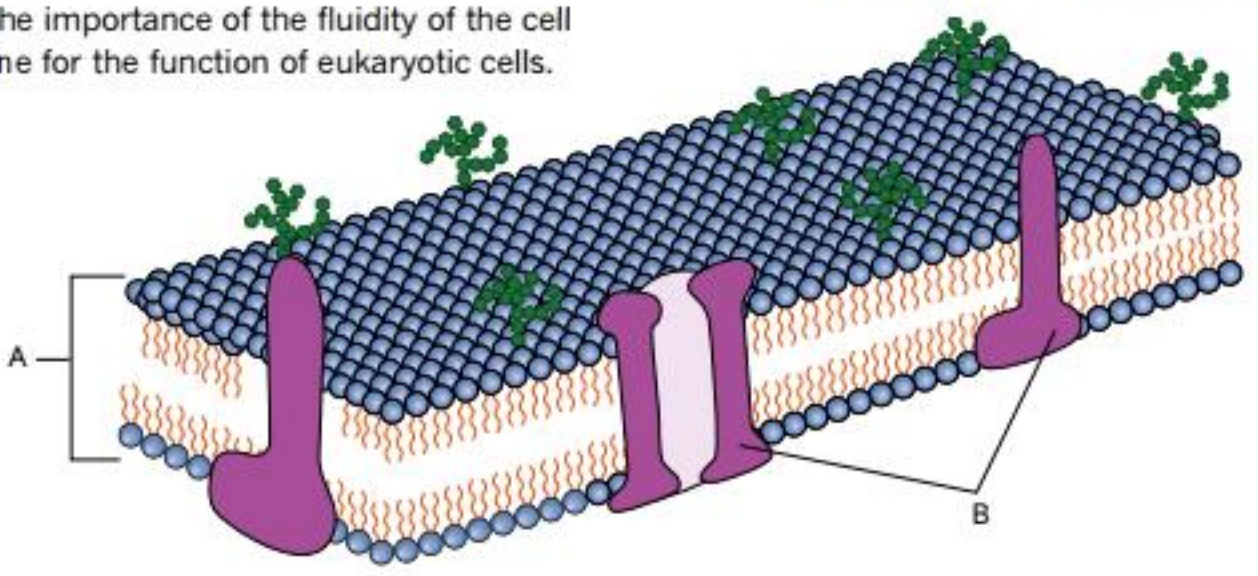
8 Why would a cell membrane containing only saturated fatty acids be very rigid?

9 Explain why protein channels are needed for cell functioning.

Analyse

10 You are asked to give a one-minute summary to the class on the fluid mosaic model of the cell membrane. Consider the key points necessary for your response and organise them appropriately to clearly explain the fluid mosaic model to your class.

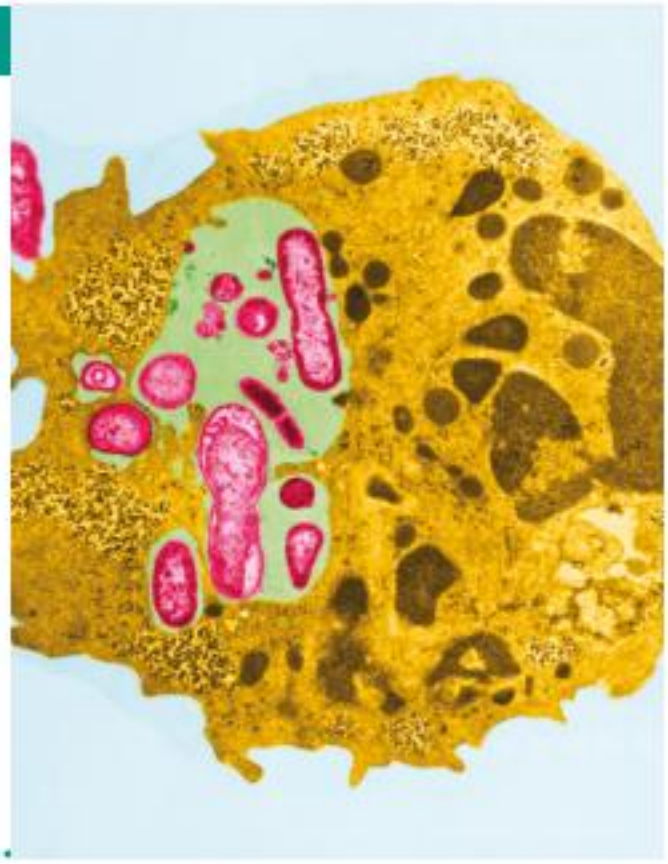
11 Marine mammals living in the Antarctic have a higher level of unsaturated fatty acids in their cell membranes than mammals living in more temperate regions. Suggest why this assists the survival of these animals.



2.4 Crossing the membrane

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand the differences between diffusion, osmosis, facilitated diffusion and active transport and name some examples of the types of substances transferred by each method
- understand the importance of surface area-to-volume ratio for cell function
- understand that endocytosis is a form of active transport that usually moves large polar molecules that cannot pass through the hydrophobic cell membrane into the cell
- predict the direction of movement of materials across the cell membrane on the basis of factors such as concentration, physical and chemical nature of the materials
- explain how the size of a cell is limited by the relationship between surface area-to-volume ratio and the rate of diffusion
- calculate surface area-to-volume ratios
- conduct an investigation to compare the efficiency of movement of materials that have different surface area-to-volume ratios.



In the previous module, you learnt about the composition of the cell membrane, and that one of its main characteristics is exchanges of molecules between the cytoplasm and the external environment of the cell. Small molecules and water are constantly transported across the cell membrane in both directions. Depending on their size and polarity, molecules diffuse between the phospholipid molecules or pass through channels formed by proteins embedded within the membrane. For larger molecules such as proteins and polysaccharides, bulk transport across the cell membrane is used. In this section, you will learn about the selective permeability of the cell membrane. You will also explore the various methods employed to control the exchange of molecules, including diffusion, facilitated diffusion, osmosis, active transport and bulk transport by endocytosis or exocytosis.

CELL MEMBRANE PERMEABILITY

To maintain the composition of the intracellular fluid, cells must control which molecules move into and out of the cell across the cell membrane. Cells can regulate what moves in and out of the cell because they have **semipermeable membranes** and transmembrane proteins. The phospholipid bilayer acts like a molecular sieve, controlling what moves between the intracellular and extracellular environments. As Figure 2.4.1 shows, many different types of molecules can move across cell membranes, and they do so in different ways, depending on their properties, such as size, charge, polarity and solubility (Table 2.4.1, page 38).

Semipermeable membranes allow solvent molecules to pass through but prevent some of the solute molecules from doing so.

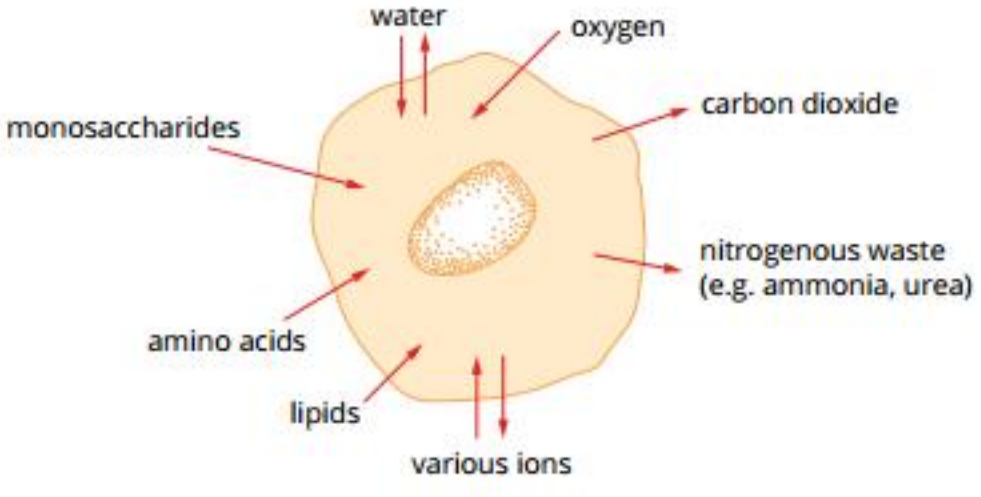


FIGURE 2.4.1 Cells exchange many substances with their environment across the cell membrane.

TABLE 2.4.1 The cell membrane's permeability to different molecules

Molecule or ion	Examples	Permeability of membrane to the molecule or ion
Small uncharged molecule	oxygen, carbon dioxide	permeable
Lipid-soluble, non-polar molecule	alcohol, chloroform, steroids	permeable
Small polar molecule	water, urea	permeable or semipermeable
Small ion	potassium ion (K^+) sodium ion (Na^+), chloride ion (Cl^-)	impermeable (molecule passes through protein channels)
Large, polar, water-soluble molecule	amino acid, glucose	impermeable (molecule passes through protein channels)

A solute is a substance dissolved in another substance, known as the solvent.

Permeable membranes are not selective in what molecules pass through them. All solutes and the solvent can pass easily across permeable membranes. Membranes are said to be 'semipermeable' or 'selectively permeable' when they allow some particles or solutes and the solvent to pass through the phospholipid part of the cell membrane, but not other solutes. Figure 2.4.2 illustrates the movement of particles across permeable and semipermeable membranes.

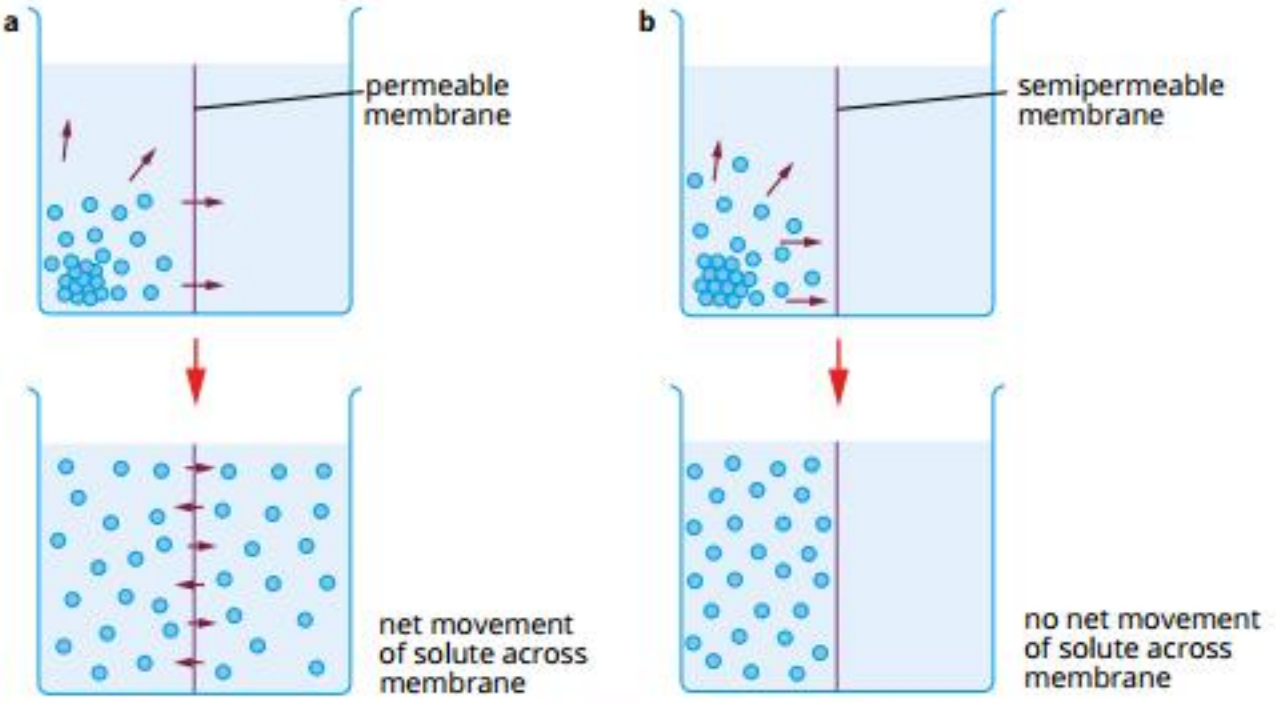


FIGURE 2.4.2 (a) When a membrane is permeable, to the solute and the solvent, the solute particles cross the membrane down the concentration gradient. (b) If the membrane is impermeable to the solute, then only the solvent can cross the membrane and the solute is prevented from diffusing through the membrane.

Because of their hydrophobic lipid nature, cell membranes are permeable to small, uncharged molecules and lipid-soluble molecules. In other words, small, uncharged molecules and lipid-soluble molecules can move freely through the phospholipid bilayer. However, the lipid nature of cell membranes makes them impermeable to:

- most water-soluble molecules
- ions (atoms or groups of atoms with an overall positive or negative charge)
- polar molecules (molecules with charged regions but no overall charge).

Membranes that do not allow substances to diffuse across them are referred to as **impermeable membranes**. Molecules that cannot diffuse across the phospholipid bilayer may enter and exit the cell through specific protein channels in the cell membrane, which are shown in Figure 2.4.3.

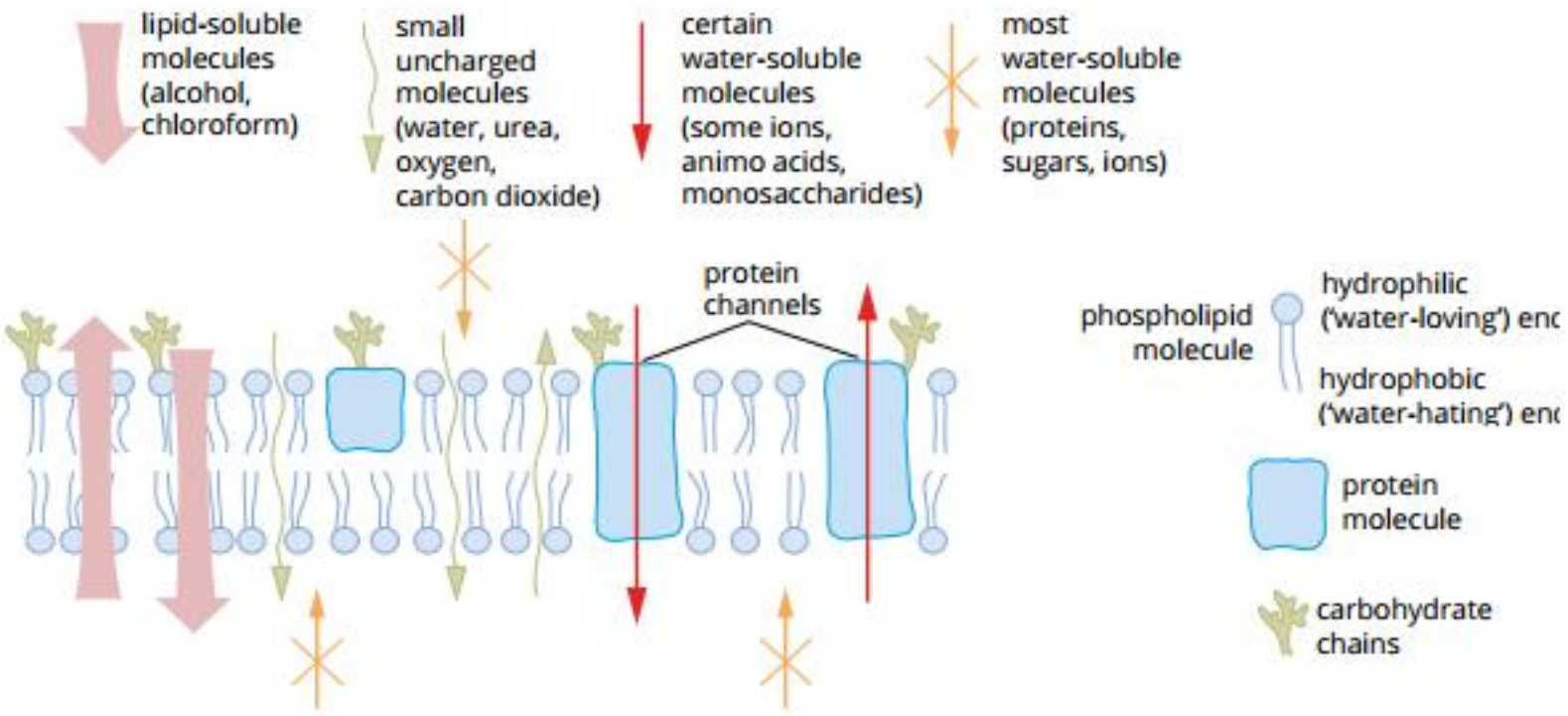

lipid-soluble molecules (alcohol, chloroform)
small uncharged molecules (water, urea, oxygen, carbon dioxide)
certain water-soluble molecules (some ions, animo acids, monosaccharides)
most water-soluble molecules (proteins, sugars, ions)
protein channels
phospholipid molecule
hydrophilic ('water-loving') end
hydrophobic ('water-hating') end
protein molecule
carbohydrate chains

FIGURE 2.4.3 If the cell membrane is not permeable to molecules, protein channels must assist the molecules to cross the membrane.

PASSIVE TRANSPORT

Passive transport is the movement of molecules without the expenditure of energy. The three types of passive transport across membranes are: diffusion, facilitated diffusion and osmosis.

Diffusion

Particles in a solution move from an area of high concentration to an area of low concentration. This process is called diffusion and is shown in Figure 2.4.4.

Particles are always in constant random motion. The random motion is a result of the kinetic energy (energy of movement) of the molecules or ions and results in many collisions of the particles. Because there are many particles colliding with each other during this process, the overall movement of particles is very slow.

Solute molecules can diffuse across a membrane only if the membrane is permeable to them. There is a constant movement of solute molecules backwards and forwards across the membrane. If the solute concentration is higher on one side of the membrane than the other, more solute molecules cross from the area of higher concentration to the area of lower concentration (i.e. down its **concentration gradient**), as you can see on the left side of Figure 2.4.5. However, if the concentration of solute molecules is the same on both sides of the membrane, there is always about the same number moving across in either direction. That is, there is no net movement from one side to the other.

If the membrane is semipermeable (i.e. it is impermeable to some molecules) there is no movement of those molecules from the area of higher concentration to the area of lower concentration, as you can see on the right side of Figure 2.4.5.

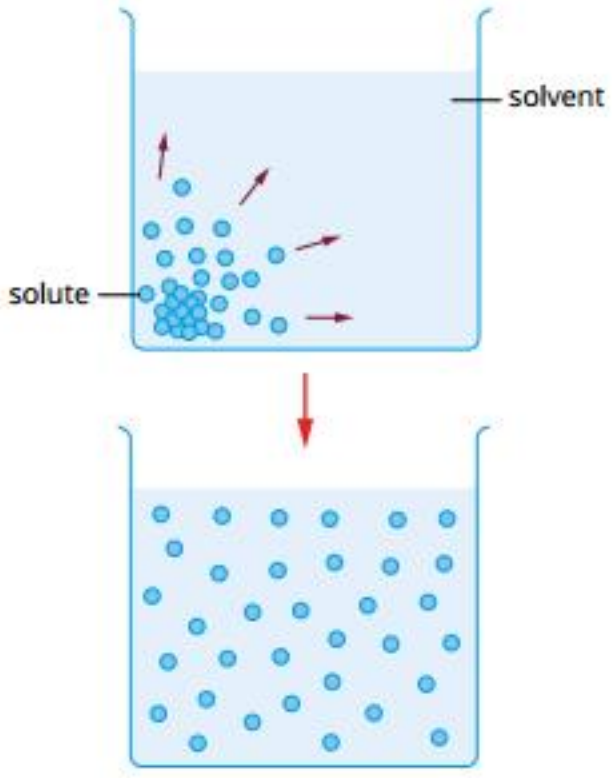


FIGURE 2.4.4 Diffusion results in the random dispersal of solute molecules throughout a solvent.

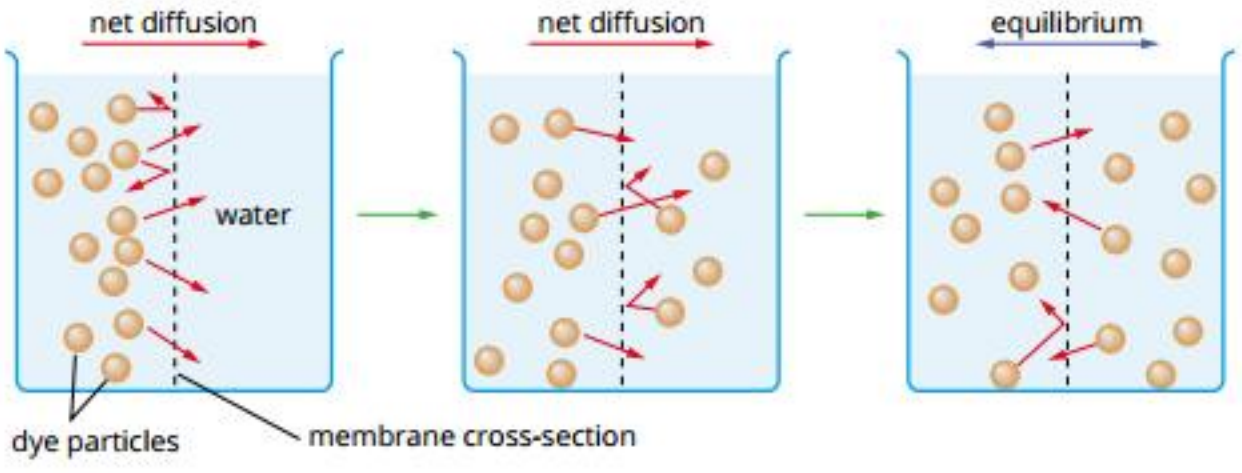


FIGURE 2.4.5 The solute particles are able to diffuse across the semipermeable membrane from high concentration (on the left) to low concentration. When equilibrium is reached, solute particles continue to randomly diffuse from both sides of the membrane but there is no change to the overall concentration. Dynamic equilibrium has been established.

i Diffusion is the passive net movement of molecules from a region where they are in high concentration to a region where they are in low concentration.

Diffusion can be seen when a drop of ink (the **solute**) is placed in a jar of still water (the **solvent**). The dye particles in the ink move randomly through the water until the colour is homogenous (evenly spread). In other words, the solute particles move from an area of high solute concentration (the drop of ink) to the areas of low solute concentration (the rest of the jar). The solute particles are said to have moved down the concentration gradient.

Diffusion is called a passive process because it does not require additional energy from outside the system (in the form of ATP [adenosine triphosphate]). It occurs only because there is a concentration gradient.

Factors affecting the rate of diffusion

The three main factors that affect the rate of diffusion across a membrane are:

- concentration—the greater the difference in concentration, the higher the rate of diffusion. When the concentration is equal on both sides of the membrane, the net diffusion is zero, even at high temperatures
- temperature—the higher the temperature, the higher the rate of diffusion. Increasing the temperature increases the speed at which molecules move
- particle size—the smaller the particles, the higher the rate of diffusion through a membrane.

Facilitated diffusion

The phospholipid bilayer of the membrane is impermeable to certain particles, including ions and large polar molecules. However, certain proteins in the membrane allow for the diffusion of these particles into and out of the cell. Because the diffusion of these molecules is assisted by proteins and does not require any additional energy to be added to the system, the process is called **facilitated diffusion**.

In facilitated diffusion:

- the membrane transport proteins are specific for particular particles, so transport is selective; some particles are transported and others are not
- transport is more rapid than by simple diffusion
- the transport proteins can become saturated (fully occupied) as the concentration of the transported substances increases
- the transport of one particle may be inhibited by the presence of another particle that uses the same transport protein
- no additional energy is required; the particles move down their own concentration gradient.

The two main types of membrane transport proteins in facilitated diffusion are **channel proteins** and **carrier proteins**. Membrane proteins provide channels for the passage of water-soluble (polar) molecules and ions across the phospholipid bilayer. Channel proteins are specific for a substance. They do not usually bind with the molecules being transported. Channel proteins function like pores that open and close to allow the passage of specific molecules. They are mainly involved in the passage of water-soluble polar particles, such as ions. Special channel proteins, called **aquaporins**, facilitate the movement of water across the membrane.

Carrier proteins bind the molecules being transported, causing the protein to change shape (conformation), which allows specific molecules to be transported across the membrane. You can see this in Figure 2.4.6. After the molecule has crossed the membrane, the carrier protein is restored to its original shape.

Both carrier proteins and channel proteins are specific to the molecules they carry and without these proteins, active transport and facilitated diffusion of particular molecules cannot occur. A cell can regulate which substances can enter or leave the cell by controlling which carrier proteins or channel proteins are present in the membrane. The specific proteins will only be present if the genes that code for them in the nucleus are active. Different cells may have different carrier proteins and channel proteins present and thus be able to import or export different substances.

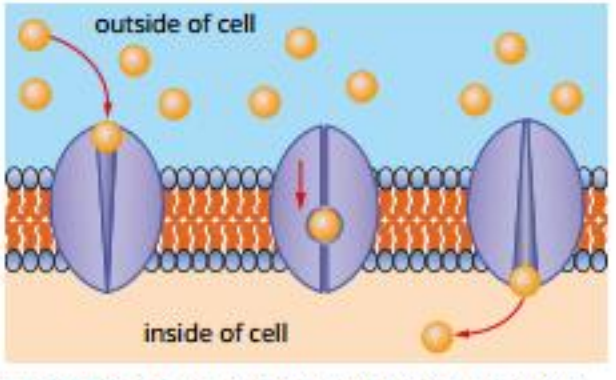


FIGURE 2.4.6 In facilitated diffusion, particles move from one side of the membrane down the concentration gradient mediated by specific carrier proteins or channel proteins.

Osmosis

Osmosis refers to the net diffusion of water molecules across a semipermeable membrane.

If a dilute and a concentrated solution are separated by a semipermeable membrane that allows the movement of free water molecules but not solute molecules, the free water molecules move across the membrane from the dilute to the concentrated solution.

In osmosis, net diffusion of water occurs through a semipermeable membrane from a dilute to a concentrated solution in an effort to achieve equilibrium. Diffusion occurs down water's concentration gradient, which is also known as the **osmotic gradient** and is shown in Figure 2.4.7. The pressure causing the water to move along this gradient is called **osmotic pressure**.

Osmosis is the movement of water from a solution of lower concentration to a solution of higher concentration.

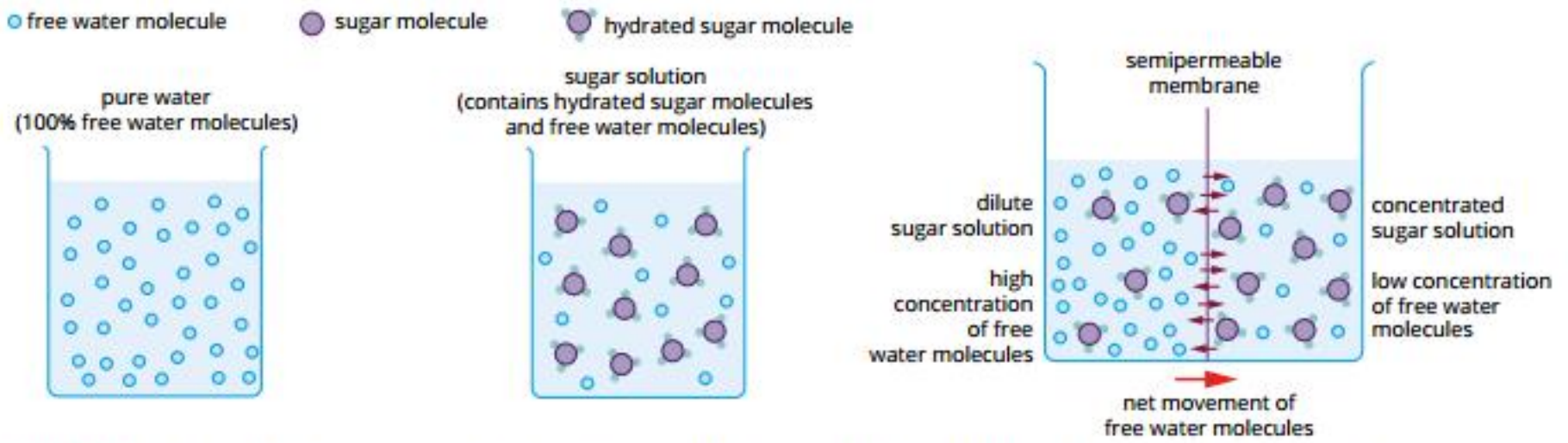


FIGURE 2.4.7 In osmosis there is a net movement of water molecules from a dilute solution through a semipermeable membrane into a concentrated solution.

Osmosis can be demonstrated by using Visking tubing (containing a strong sugar solution coloured with food dye) attached to a clear capillary tube and submerged in a beaker of water, as shown in Figure 2.4.8. Visking tubing is a synthetic semipermeable membrane made from cellulose. In the Visking tubing, water molecules bind to the sugar molecules. As a result, there are fewer free water molecules in the Visking tubing and a net movement of free water molecules into the tubing by osmosis occurs. This causes an increase in the volume of liquid in the tubing, increasing the pressure and forcing the coloured solution to rise up the capillary tube.

In osmosis, we are always comparing solute concentration between two solutions. The terms 'isotonic', 'hypertonic' and 'hypotonic' solutions are often used to describe the difference.

- Isotonic solutions: the solutions being compared have equal concentrations of solutes.
- Hypertonic solutions: the solutions with higher concentration of solute (and hence lower concentration of free water molecules).
- Hypotonic solutions: the solutions with lower concentration of solute (hence higher concentration of free water molecules).

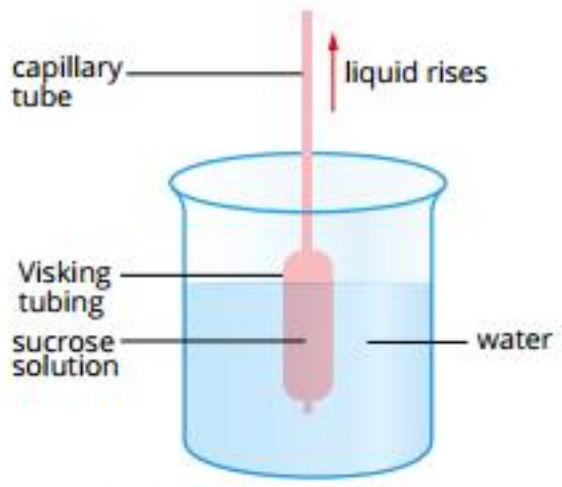


FIGURE 2.4.8 A Visking tubing apparatus for the demonstration of osmosis

Effect of osmosis on cells

The cell membrane is permeable to water, so when cells are placed in fresh water, an osmotic gradient draws water into the cells. This is because the cytosol is a concentrated solution containing many dissolved substances. In other words, the cytosol has a low concentration of water. For example, if red blood cells are placed in fresh water, the cells absorb so much water by osmosis that they swell and may eventually burst (lysis), releasing red pigment into the water, as shown in Figure 2.4.9c on page 42. Conversely, if red blood cells are placed in a solution that is more concentrated than their cytosol, water leaves the red blood cells by osmosis and causes them to shrink (crenation). This is shown in Figure 2.4.9a on page 42.

For cells with cell walls, such as plant cells and prokaryotes, the cell wall helps to maintain the cell's water balance. For example, if a plant cell loses water by osmosis, it starts to shrivel, and the cell membrane starts to pull away from the cell wall—the cell is said to have become plasmolysed (Figure 2.4.9d). However, if the plant cell absorbs water by osmosis, it swells to some extent, but the relatively inelastic cell wall prevents the cell from bursting. The cell wall expands until it exerts a pressure back on the cell, known as turgor pressure. Turgor pressure prevents further water uptake. At this point, the plant cell is turgid (Figure 2.4.9f).

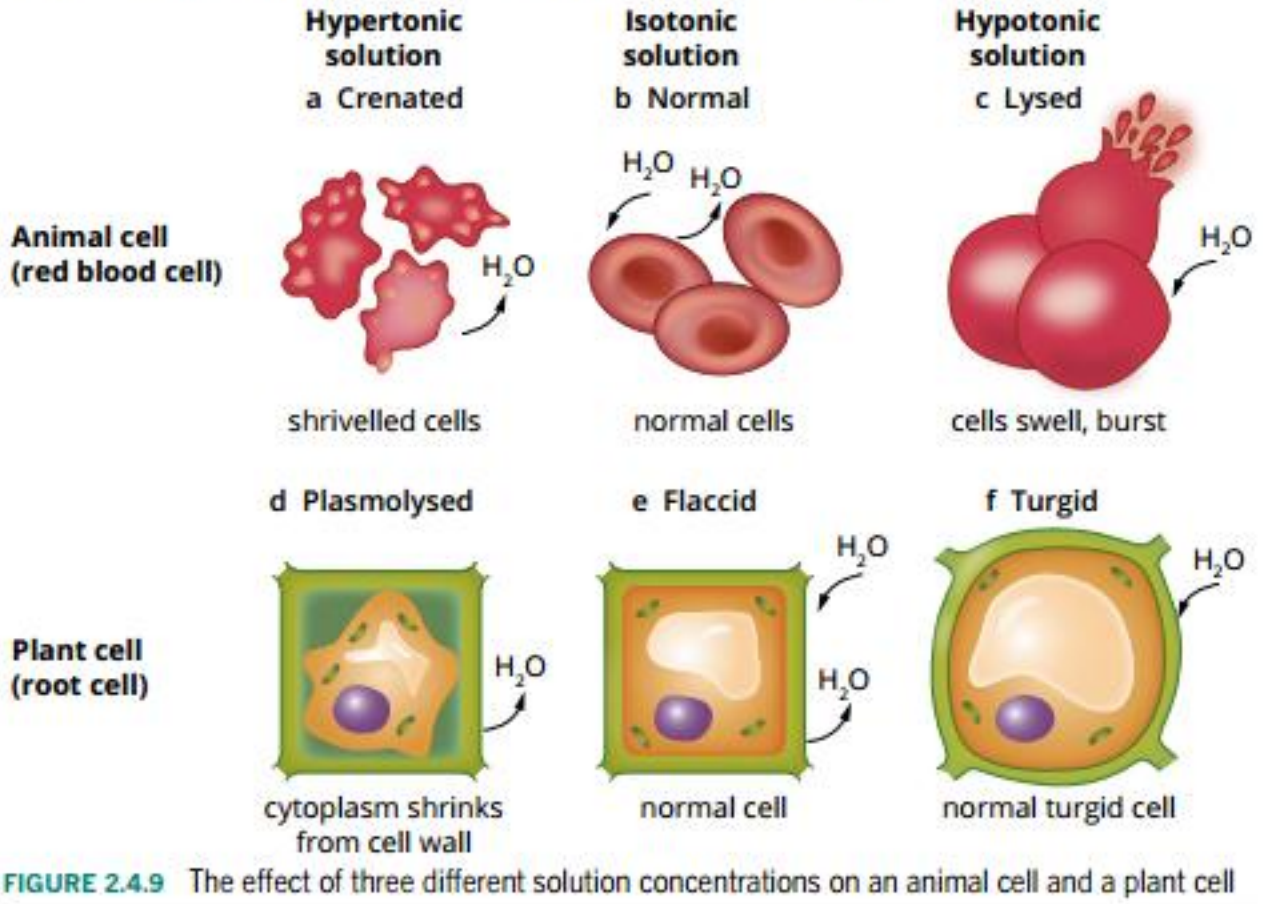


FIGURE 2.4.9 The effect of three different solution concentrations on an animal cell and a plant cell

ACTIVE TRANSPORT

Active transport requires the use of energy to move substances against a concentration gradient.

Figure 2.4.10 illustrates **active transport**. Active transport involves the use of energy by the cell to transport particles across membranes. Because active transport uses energy, it can move substances against a concentration gradient from low concentration to high concentrations. Active transport enables a cell to maintain internal concentrations of small solutes that differ from concentrations in its environment. The membrane transport proteins in active transport are all carrier proteins.

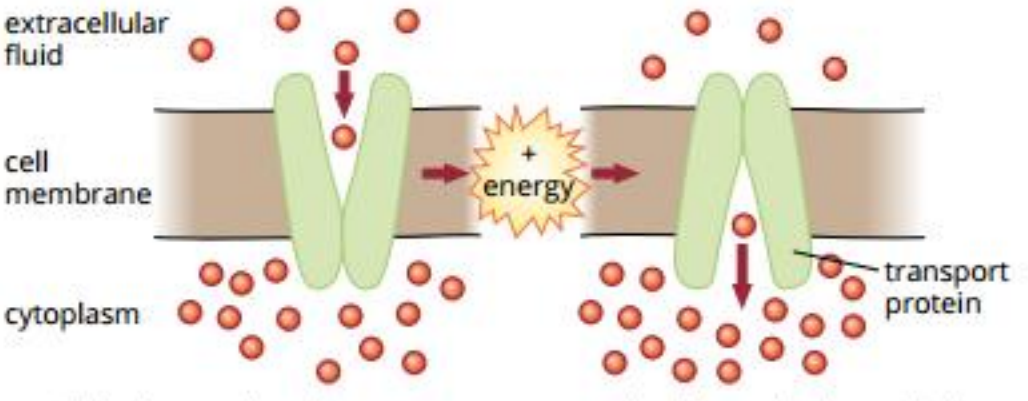


FIGURE 2.4.10 Active transport requires an energy source so that the molecules can be transported across the membrane.

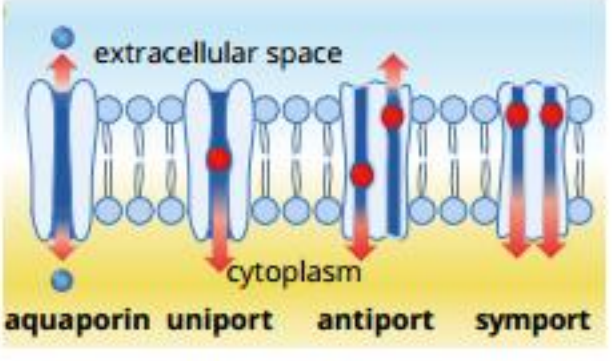


FIGURE 2.4.11 Four types of protein transport channels exist. Aquaporins mainly carry water. Uniport channels carry only one type of molecule or ion and antiport and symport channels carry more than one type of molecule or ion and can be involved in secondary active transport.

In active transport, carrier proteins use energy in the form of ATP to move substances across the cell membrane. This process uses carrier proteins, of which there are three different types: **uniporter**—which carries only one type of molecule or ion, **symporter**—which carries two different molecules or ions in the same direction, and **antiporter**—which carries two different molecules or ions in two different directions (Figure 2.4.11). The processes involved in allowing the molecules to cross the membrane involve a change in shape of the carrier protein.

These transport proteins act as pumps moving ions across the membrane, resulting in an electrochemical imbalance with one side of the membrane being positively charged while the other side is slightly negative. This electrical difference supplies energy that can then be used to move other substances across the membrane.

One of the most important pumps is the sodium/potassium (Na^+/K^+) pump. The protein carrier has binding sites for three sodium ions and two potassium ions.

The process is as follows:

- Three sodium ions inside the cell bind to the protein then an ATP molecule donates its phosphate, which binds to the protein.
- The protein changes shape, and the three sodium ions are released outside the cell.
- Two potassium ions now bind to the carrier protein, causing the shape of the protein to change back and the phosphate to be released.
- The two potassium ions are released inside the cell.
- Three sodium ions leave the cell for every two potassium ions that enter the cell, causing the electrical imbalance between inside and outside the cell.

Once the electrical imbalance is formed the electrical energy stored can be used to power the movement of other molecules across the membrane in a process called secondary active transport.

There is now a high concentration of sodium ions outside the cell and a low concentration inside the cell. If another protein transporter that allows sodium into the cell is present, sodium ions will start to move back into the cell via this route. If the carrier is a symporter, it can carry another molecule, such as glucose, as a 'passenger' into the cell while carrying in the sodium. This may move the glucose up its concentration gradient. If the carrier is an antiporter, then the 'passenger' molecule is moved out of the cell. Although no ATP is used at this stage, it is still considered active transport because it relies on the high concentration of sodium ions outside the cell, which is created using ATP.

Active transport and facilitated diffusion compared

Passive transport does not require an additional energy source. There are three types of passive transport: diffusion, facilitated diffusion and osmosis. Diffusion (Figure 2.4.12a) occurs when substances move from high to low concentrations. In facilitated diffusion (Figure 2.4.12b), substances move from high to low concentrations with help from a transport protein. Osmosis is the movement of water from high to low

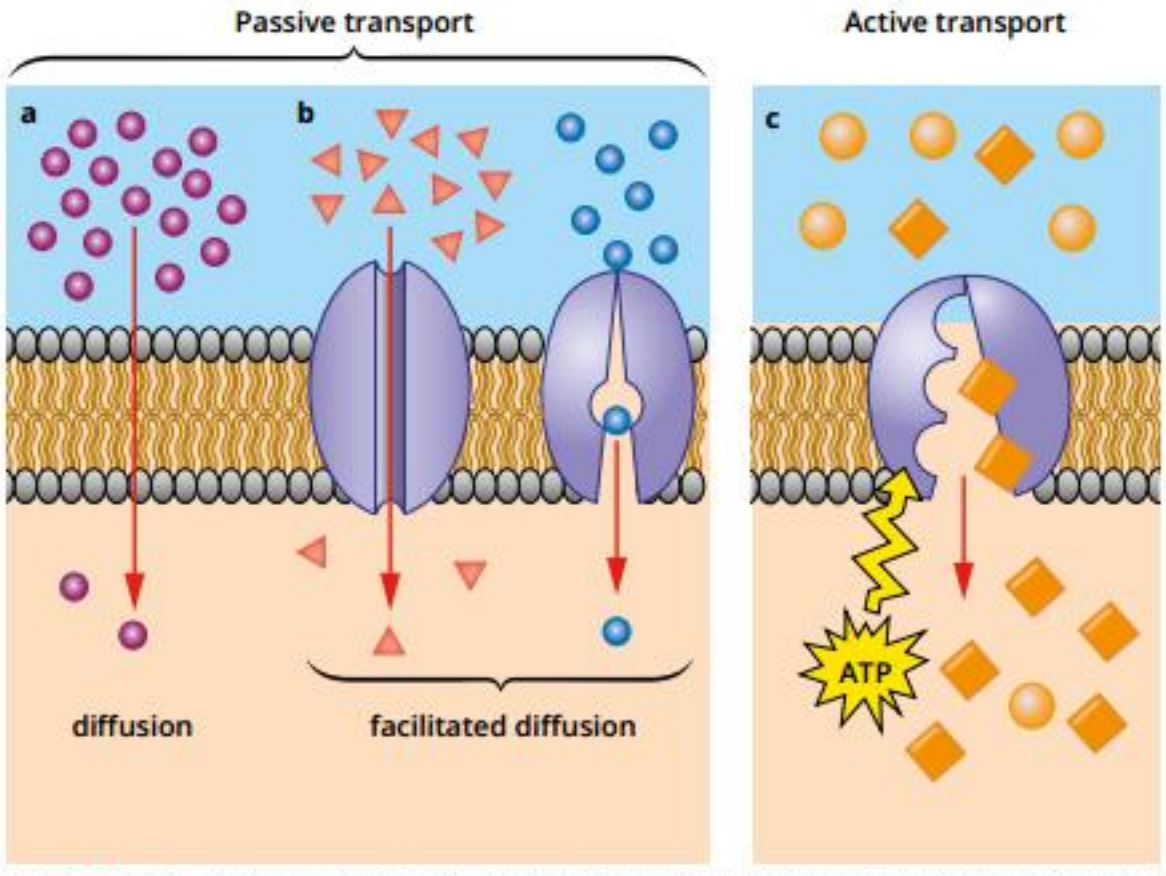


FIGURE 2.4.12 Passive transport of molecules across the membrane does not require energy. Active transport does require an energy source.

water concentrations. Active transport (Figure 2.4.12c) requires an energy source. As a result, it usually moves substances from low to high concentrations.

The comparison between passive transport and active transport can be seen in Table 2.4.2.

TABLE 2.4.2 A comparison of passive and active transport

	Passive transport			Active transport
	Simple diffusion	Osmosis	Facilitated diffusion	
Type of substance	hydrophobic molecules, small polar molecules; for example, water, oxygen, carbon dioxide	water	hydrophilic molecules; for example, calcium ions, glucose, amino acids, sodium ions	Hydrophilic molecules; for example, glucose, amino acids, sodium ions, potassium ions
Type of membrane protein required	none	none	channel protein carrier protein	Carrier protein
Direction of movement of molecules	down concentration gradient	down concentration gradient	down concentration gradient	Against concentration gradient
Energy requirement	none	none	none	Requires energy in the form of ATP

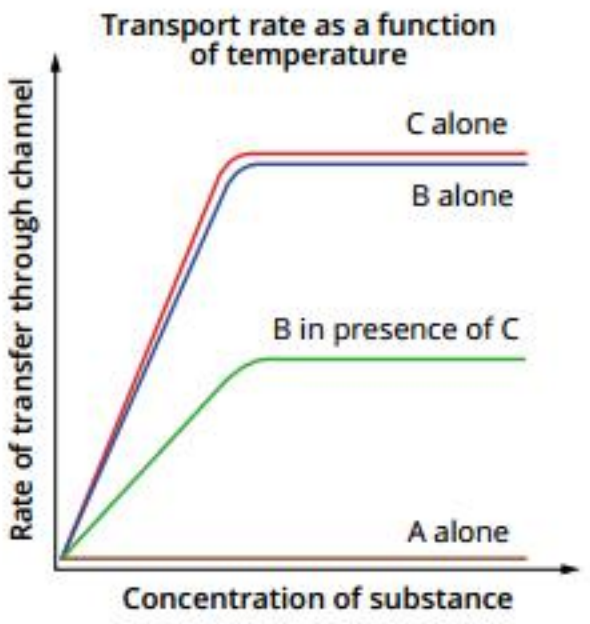


FIGURE 2.4.13 Theoretical transport rate versus concentration for the movement of three substances through a channel protein. Substances B and C are transported, but not substance A—this demonstrates selectivity. The rate of transfer of substances B and C flattens out when their concentrations reach a certain level, demonstrating saturation. The rate of transport of B is less when C is present, demonstrating competitive inhibition.

Active transport has the same properties of selectivity, saturation and competitive inhibition as facilitated diffusion because it also occurs through transport proteins (Figure 2.4.13). Selectivity means that some substances are transported but others are not. Saturation means that there is no increase in the rate of transfer when all transport proteins are open. Competitive inhibition means that one substance can inhibit the transport of another substance by using the same transport protein.

But unlike facilitated diffusion, which can occur through either channel or carrier proteins, active transport only occurs through carrier proteins. Because active transport uses energy, it can move substances against a concentration gradient (from low concentrations to high concentrations). In comparison, facilitated diffusion uses no energy, so it can only move substances down a concentration gradient.

In different situations, either facilitated diffusion or active transport may be used to transport a particular molecule. Whether a cell uses facilitated diffusion or active transport depends on the specific needs of the cell.

For example, glucose is actively transported from the gut into epithelial cells lining the gut so it can enter the bloodstream. This process is regulated by hormones, principally insulin and glucagon. If gut glucose levels are high, blood glucose levels increase. If gut glucose levels are low, active transport makes sure that the little glucose that is in the gut gets pumped into the epithelium from where it can move to blood through facilitated diffusion.

In contrast, red blood cells move glucose by facilitated diffusion. This makes sense because glucose concentration in the blood is usually maintained within a narrow range. In addition, cells convert glucose into other chemicals as soon as it enters the cell, keeping the intracellular concentration of glucose lower than the blood concentration of glucose.

BULK TRANSPORT

Large polar molecules and other substances that cannot pass through the hydrophobic cell membrane can enter or exit the cell via bulk transport. Bulk transport includes exocytosis and endocytosis (Figure 2.4.14). Both exocytosis and endocytosis are forms of active transport because they require energy.

Exocytosis and endocytosis

Figure 2.4.14 illustrates how cells transport large molecules into and out of the cell. **Exocytosis** is the movement of substances out of the cell, from the cytoplasm to the extracellular fluid. A transport **vesicle**, which may contain wastes or substances needed for secretion (e.g. digestive enzymes, hormones or neurotransmitters), fuses with the cell membrane and the junction then breaks down, releasing the enclosed materials. Transport vesicles are made of the same lipid bilayer as the cell membrane and are thus easily able to fuse with it. It should be noted as seen in the Figure 2.4.14 that the contents of the vesicle exit the cell, but the vesicle becomes part of the cell membrane. Unicellular heterotrophs such as amoebas remove digestive wastes in this way.

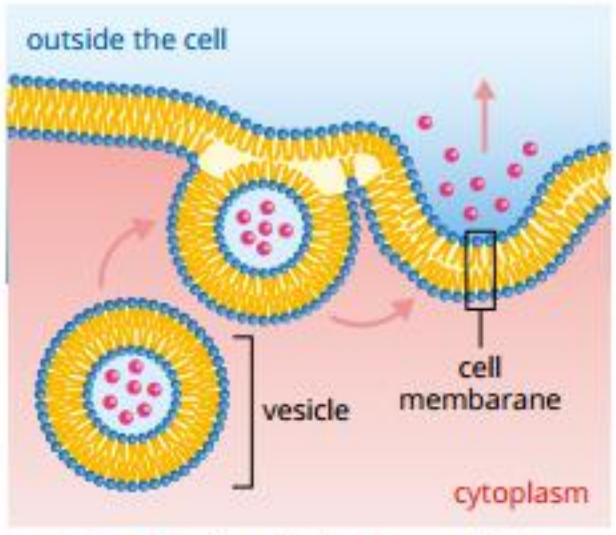


FIGURE 2.4.14 Exocytosis moves substances out of the cell using transport vesicles. The vesicles are made of a phospholipid bilayer and easily fuse with the cell membrane, releasing its contents out of the cell.

Endocytosis is the movement of substances into the cell, from the extracellular fluid into the cytoplasm. Particles near the cell membrane are enclosed by the membrane, which then pinches off to form a vesicle enclosing the particles, as seen in Figure 2.4.15. In eukaryotes, this vesicle may become fused with a lysosome so that its contents can be digested for use by the cell.

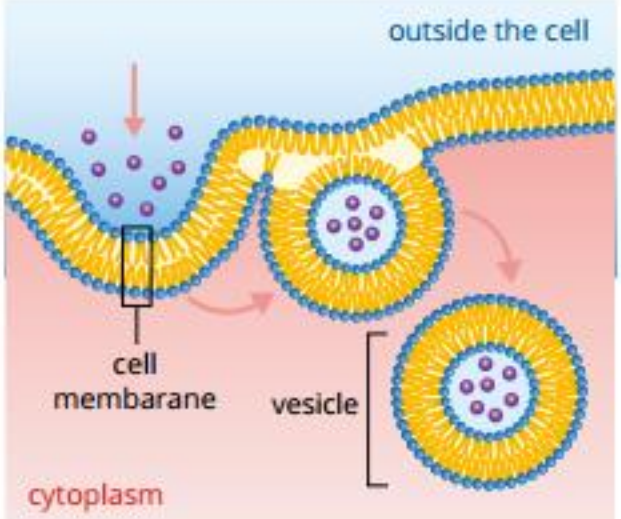


FIGURE 2.4.15 Endocytosis moves substances into the cell by forming vesicles from the cell membrane around particles.

The process varies slightly depending on the substance to be absorbed. The two forms of endocytosis are pinocytosis and phagocytosis (Figure 2.4.16). **Pinocytosis** is the entry of extracellular fluid and substances such as proteins and sugars that are carried in it. **Phagocytosis** is the entry of large particles such as bacteria and cell debris to be broken down as an immune response. In phagocytosis, pseudopodia surround the particle whereas in pinocytosis a pocket forms around the liquid. Pseudopodia are formed around solid materials but not around liquids. Stimulation of receptors on the cell surface causes the formation of the endocytic vesicle.

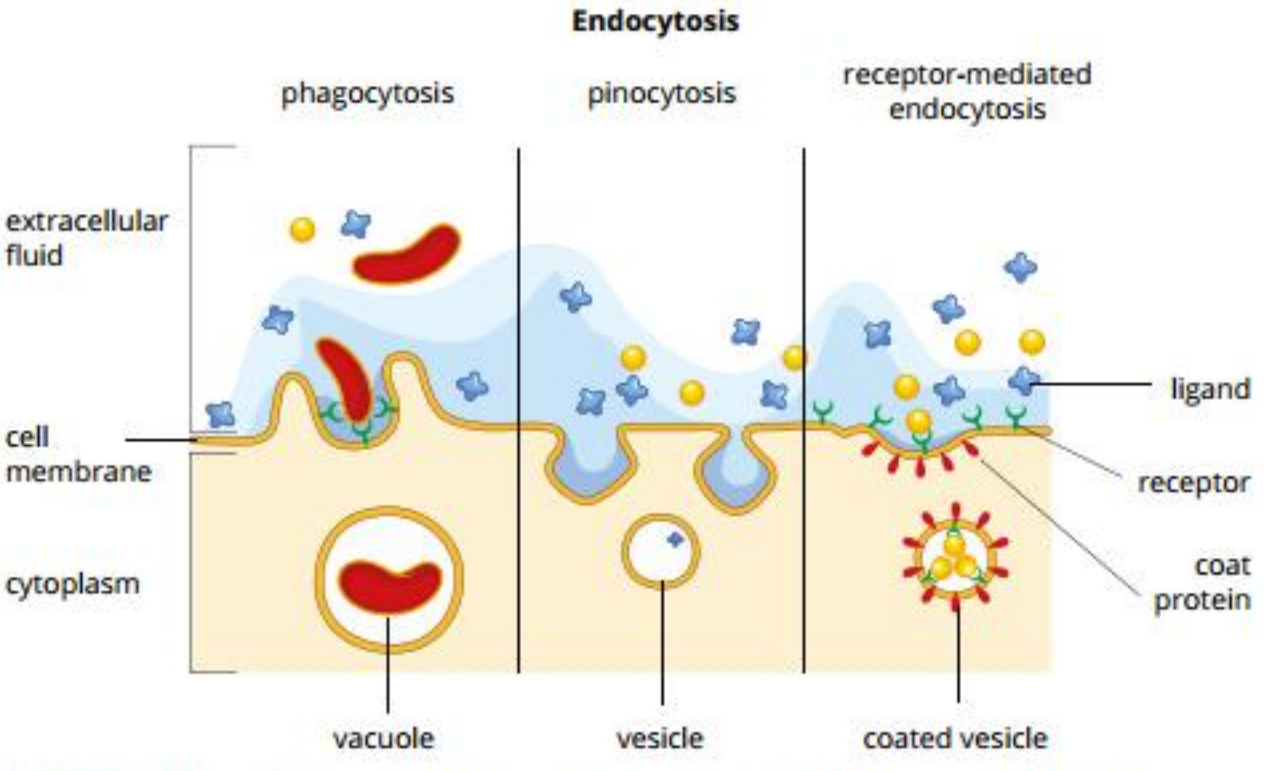


FIGURE 2.4.16 Substances move into the cell via phagocytosis or pinocytosis (the two forms of endocytosis).

Most endocytosis is receptor mediated. Receptors are proteins on the external surface of the cell membrane whose shape allows them to attach to a specific molecule (Figure 2.4.16). The attachment of the molecule to the receptor triggers the changes in the cell membrane to form the vesicle around the particle.

SURFACE AREA-TO-VOLUME RATIO AND CELL SIZE

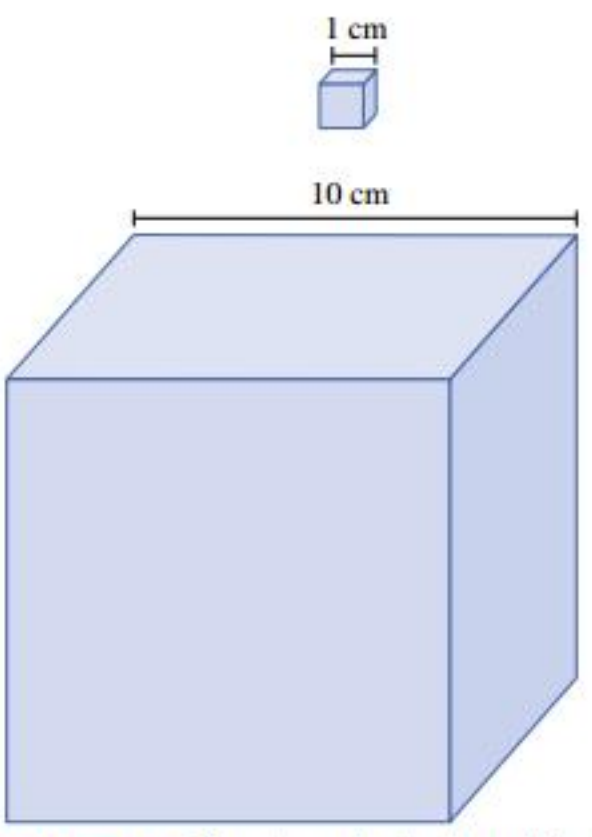
All cells must exchange nutrients and wastes with their environment via the cell membrane. In addition, enzymes that are bound to the cell membrane catalyse many important cellular processes.

The surface area of the cell membrane around a cell affects the rate of exchange that is possible between the cell and its environment and can affect certain processes catalysed by membrane-bound enzymes.

Larger cells have greater metabolic needs, so they need to exchange more nutrients and waste with their environment. However, because of the surface area-to-volume relationship, they do not have a proportionally larger surface area of cell membrane for this exchange to take place. Thus the small size of the cells help maximise their efficiency in exchanging matter with their environment.

Surface area versus volume

The relationship between surface area and volume can be explained using cubes, as shown in Figure 2.4.17. A cube with a side length of 1 cm has a surface area of 6 cm^2 and a volume of 1 cm^3. The surface area-to-volume ratio of a 1 cm cube is thus 6:1 (or 6). A cube with a side length of 10 cm has a surface area of 600 cm^2 and a volume of 1000 cm^3. A 10 cm cube thus has a surface area-to-volume ratio of 600:1000, or 0.6. Comparing these two cubes, it can be observed that, while the volume of the bigger cube is 1000 times larger than the volume of the smaller cube, its surface area is only 100 times larger.



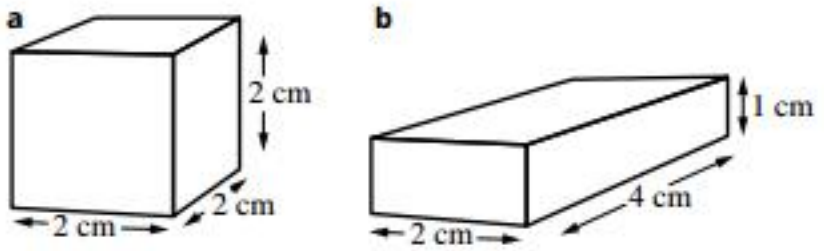
Side length	1 cm	10 cm
Surface area	6 cm^2	600 cm^2
Volume	1 cm^3	1000 cm^3
Surface area-to-volume ratio	6	0.6

FIGURE 2.4.17 Two cubes, showing the relationship between surface area and volume

Worked example 2.4.1

SURFACE AREA AND VOLUME

Consider the two objects shown below. Both objects have a volume of 8 cm^3. Which shape has the greater surface area-to-volume ratio?

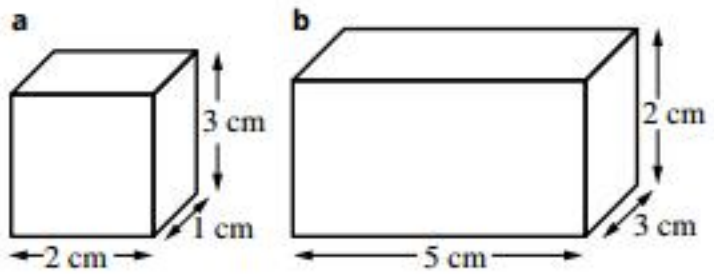


Thinking	Working
Look at shape A. Identify the dimensions of the cube.	Shape A: 6 sides, each with width of 2 cm and height of 2 cm
Calculate the surface area of shape A, given that there are six sides, each with the same area.	Surface area $= 6 \times 2 \times 2$ $= 24\ cm^2$
Calculate the surface area (SA)-to-volume (V) ratio of shape A.	$\frac{SA}{V} = \frac{24}{8}$ $= 3$
Look at shape B. Identify the dimensions of the cube.	Shape B: 2 sides, each with width of 2 cm and height of 1 cm 2 sides, each with width of 4 cm and height of 1 cm 2 sides, each with width of 2 cm and height of 4 cm
Calculate the surface area of shape B, given that there are six sides with three different areas.	Surface area $= (2 \times 2 \times 1) + (2 \times 4 \times 1) + (2 \times 2 \times 4)$ $= 4 + 8 + 16$ $= 28\ cm^2$
Calculate the surface area-to-volume ratio of shape B.	$\frac{SA}{V} = \frac{28}{8}$ $= 3.5$
Compare the surface area-to-volume ratios.	Therefore, shape B has the greater surface area-to-volume ratio.

➤ Try yourself 2.4.1

SURFACE AREA AND VOLUME

Consider the two objects shown below. Which shape has the greater surface area-to-volume ratio?



2.4 Review

SUMMARY

- The lipid bilayer of membranes is impermeable to most water-soluble molecules, ions and polar molecules. These substances can only pass through protein channels.
- Lipid-soluble substances can diffuse through the phospholipid bilayer.
- Diffusion is the passive movement of solute molecules along a concentration gradient, from a region of high solute concentration to a region of low solute concentration.
- There are three types of diffusion across cell membranes: simple, facilitated and osmosis.
- Simple diffusion involves solutes to which the membrane is permeable, including lipid-soluble substances, small molecules and water molecules. The rate of diffusion is affected by concentration, temperature and particle size.
- Facilitated diffusion is through selective channels in membranes that permit or enhance the passive movement of particular ions and molecules down their own concentration gradient. Facilitated diffusion generally occurs at a more rapid rate than simple diffusion.
- Osmosis is the net diffusion of water across a semipermeable membrane down its own concentration gradient called the osmotic gradient (i.e. from a low solute concentration to a high solute concentration).
- In active transport, energy is expended to move substances across cell membranes through protein channels against their concentration gradient.
  - Protein channel mediated active transport and facilitated diffusion require the presence of suitable protein channels.
- Exocytosis (moving substances out of the cell) and endocytosis (moving substances into the cell) are forms of active transport involving vesicles that fuse with the cell membrane. These forms of active transport are generally used to transport larger molecules in bulk.
- A large object has a smaller surface area-to-volume ratio than a small object with the same shape.

KEY QUESTIONS

Describe

1 Complete the following table by recalling whether the phospholipid bilayer is permeable, semipermeable or impermeable to each substance described.

Substance	Examples	Permeability
small uncharged molecule	oxygen, carbon dioxide	
lipid-soluble, non-polar molecule	alcohol, chloroform, steroids	
small, polar molecule	water, urea	
small ion	potassium ion (K^+), sodium ion (Na^+), chloride ion (Cl^-)	
large, polar, water-soluble molecule	amino acid, glucose	

2 Define 'selective permeability'.

3 List the factors that increase the rate of diffusion.

4 When concentrations of solutions on both sides of a permeable membrane are in equilibrium, there is no net movement of particles. Explain this statement.

5 What are the three kinds of carrier proteins and what do they do?

6 How do active and passive transport differ?

Apply

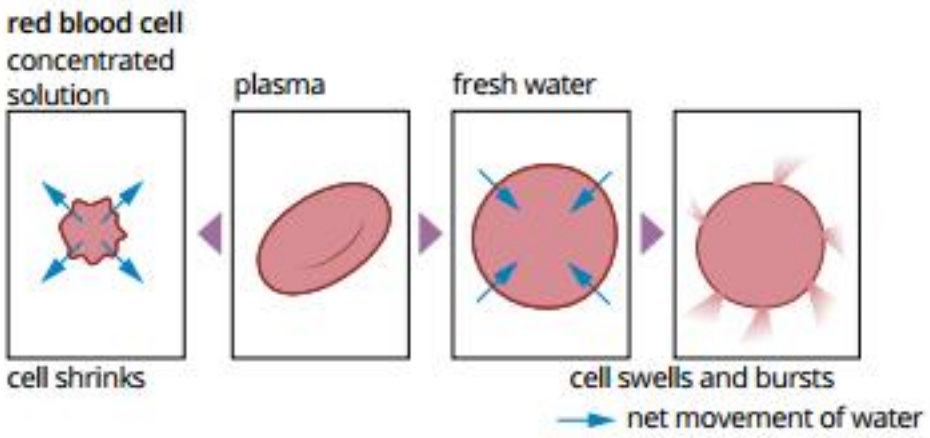
7 Osmosis is a special kind of diffusion. Define osmosis. Draw a diagram to illustrate your answer.

8 Explain how active transport is different from diffusion.

9 Consider the red blood cells in the illustrations below. The arrows indicate the net direction of water movement. Applying your understanding of osmosis, describe how red blood cells would:

a shrink

b swell and burst.



10 Explain why the cell membrane is permeable to chloroform (and equivalent chemicals).

11 Explain how the cell regulates the entry and exit of molecules and ions moving through protein channels across the cell membrane.

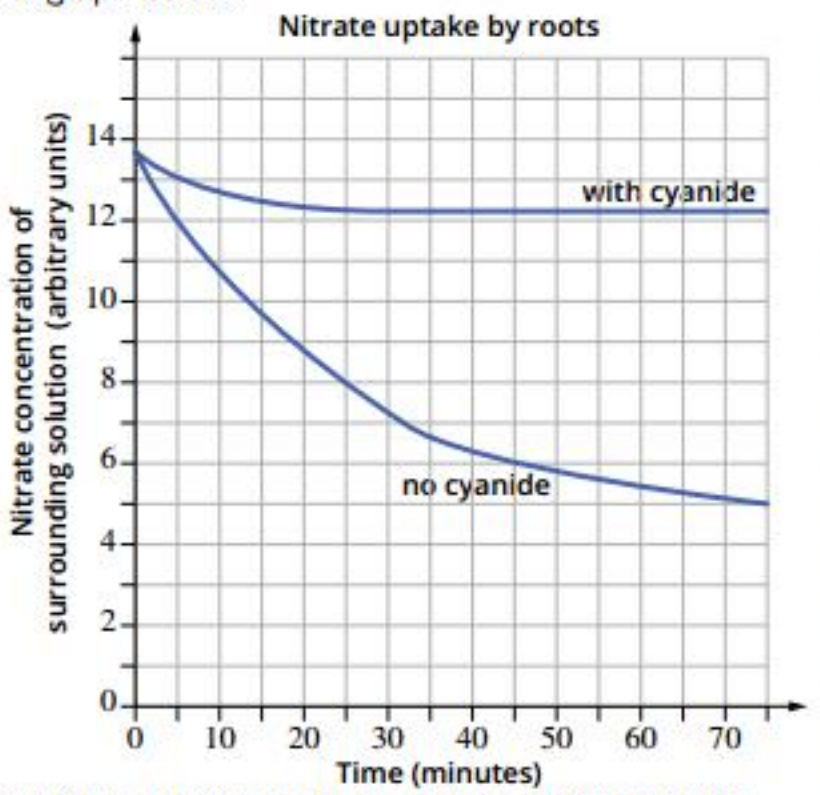
12 Determine why a large surface area-to-volume ratio is important in exchanging materials between cells and the external environment.

Analyse

13 Compare endocytosis, exocytosis, phagocytosis and pinocytosis. Describe where in your body you would expect each process to occur.

14 Compare simple and facilitated diffusion. Include an example of each.

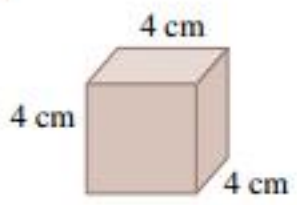
15 Plants use inorganic nitrate (NO_3^-) absorbed from the soil to make proteins. An experiment was undertaken to investigate how the nitrate enters the plant. Two groups of root cells were bathed in solutions containing nitrate ions. One of the solutions also contained cyanide, a chemical that inhibits cellular respiration. The results of the experiment are shown in the graph below.



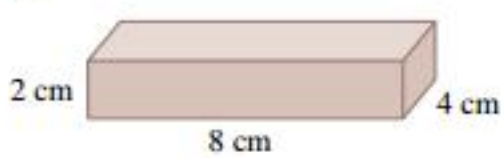
Explain the difference in the uptake of nitrate in the two situations.

16 Consider the two objects shown below which have the same volume of 64 cm^3. Determine which shape has the greater surface area-to-volume ratio.

a



b



17 Determine whether a round cell of 6 mm diameter will absorb substances via diffusion at a greater rate than a round cell of 3 mm diameter.

Chapter review

02

KEY TERMS

active transport
amino acid
antiporter
aquaporin
archaea
bacteria (sing. bacterium)
biogenesis
carrier protein
cell
cell compartmentalisation
cell membrane
cell wall
channel protein
chloroplast
cholesterol
chromosome
concentration gradient
cytology
cytoplasm
cytoskeleton
cytosol
diffusion
DNA (deoxyribonucleic acid)
endocytosis
endoplasmic reticulum
eukaryotic cell
exocytosis
extracellular fluid
extremophile
facilitated diffusion
genophore
glycolipid
glycoprotein
Golgi apparatus
grana (sing. granum)
hydrophobic
hydrophilic
impermeable membrane
inorganic compound
integral protein
intracellular fluid
lamellae (sing. lamella)
lysosome
mitochondria (sing. mitochondrion)
mRNA (messenger RNA)
murein
nucleoid
nucleus
nucleolus
organelle
organic compound
osmosis
osmotic gradient
osmotic pressure
passive transport
peripheral protein
phagocytosis
phospholipid
pinocytosis
plasmid
prokaryotic cell
protein
ribosome
RNA (ribonucleic acid)
rough endoplasmic reticulum
rRNA (ribosomal RNA)
semipermeable membrane
solute
solvent
smooth endoplasmic reticulum
stroma
symporter
taxonomy
thylakoid
tonoplast
transmembrane protein
uniporter
vacuole
vesicle

KEY QUESTIONS

Describe

1 The cell theory states that:
   - **A** all organisms are made up of cells.
   - **B** all cells arise from pre-existing cells.
   - **C** the cell is the smallest functional unit of living things.
   - **D** all of the above.

2 Why were prokaryotes divided into two domains?

3 Select the statement that accurately describes eukaryotic cells.
   - **A** Eukaryotic cells have circular chromosomes and membrane-bound organelles, and some also have cell walls.
   - **B** Eukaryotic cells have linear chromosomes but not membrane-bound organelles, and some have cell walls.
   - **C** Eukaryotic cells have linear chromosomes and membrane-bound organelles, and some also have cell walls.
   - **D** Eukaryotic cells have linear chromosomes and membrane-bound organelles, but not cell walls.

4 Identify which of the following is/are never found in prokaryotic cells.
   - **A** DNA
   - **B** mitochondria
   - **C** cytosol
   - **D** cell wall

5 List three features that distinguish prokaryotic from eukaryotic cells.

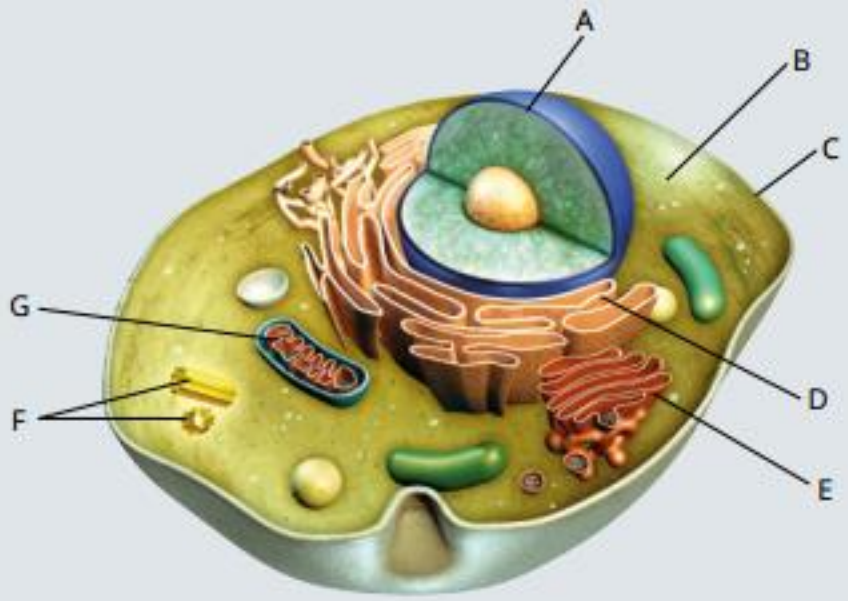
6 Draw and prepare a table to summarise the major functions of phospholipids, cholesterol, glycolipids, glycoproteins and proteins in cell surface membranes.

7 The sodium/potassium ion pump moves ions across the cell membrane. What type of protein channel does it use?

8 Copy and complete the table below by indicating if the organelles are present or absent in each type of cell.

Type of cell	Organelle					
	mitochondrion	chloroplast	endoplasmic reticulum	ribosome	nucleus	Golgi apparatus
Animal kidney cell						
Plant leaf palisade cell						
Plant root cell						
Cyanobacterium						
Achaean						
Fungal cell						

9 Label the parts of the animal cell in this diagram.



10 Many single-celled organisms such as *Amoeba* feed by a process in which the cell membrane engulfs solid food particles to form a food vacuole. This process is called:

A phagocytosis.
B active transport.
C pinocytosis.
D osmosis.

11 The organelle on which proteins are assembled is called the:

A nucleus.
B endoplasmic reticulum.
C Golgi apparatus.
D ribosome.

Apply

12 Explain which type of microscope would be best for the study of:

a changes in a white blood cell
b details of surface texture of a hair.

13 Identify whether this cell is from an animal or a plant. Give reasons for your choice.

14 Summarise the properties of archaean cell walls that allow them to be extremophiles.

15 Explain how the compartmental organisation of a eukaryotic cell contributes to its biochemical functioning.

16 According to the fluid mosaic model of membrane structure, proteins of the membrane are mostly:

A spread in a continuous layer over the inner and outer surfaces of the membrane.
B confined to the hydrophobic interior of the membrane.
C embedded in a lipid bilayer.
D randomly orientated in the membrane, with no fixed inside–outside polarity.
E free to depart from the fluid membrane and dissolve in the surrounding solution.

17 Identify which of the following factors would tend to increase cell membrane fluidity:

- **A** a greater proportion of unsaturated phospholipids
- **B** a greater proportion of saturated phospholipids
- **C** a lower temperature
- **D** a relatively high protein content in the membrane
- **E** a greater proportion of relatively large glycolipids compared with lipids having smaller molecular masses

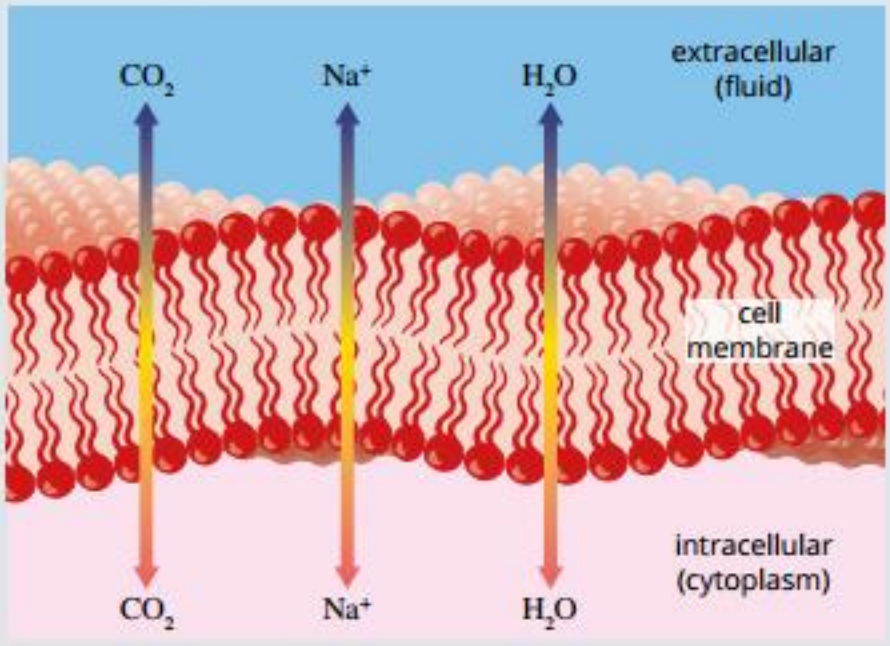
18 Explain why the phospholipid heads of the cell membrane are always pointed towards the cytosol and extracellular fluid, whereas the 'tails' are always orientated toward the middle of the membrane.

19 Describe the two types of proteins used in facilitated diffusion.

20 A thyroid gland cell was examined using a light microscope.

- **a** What organelles are likely to be visible?
- **b** The same tissue was examined using a transmission electron microscope. What differences would become visible?

21 The diagram below contains a significant error. Explain what is wrong with it.

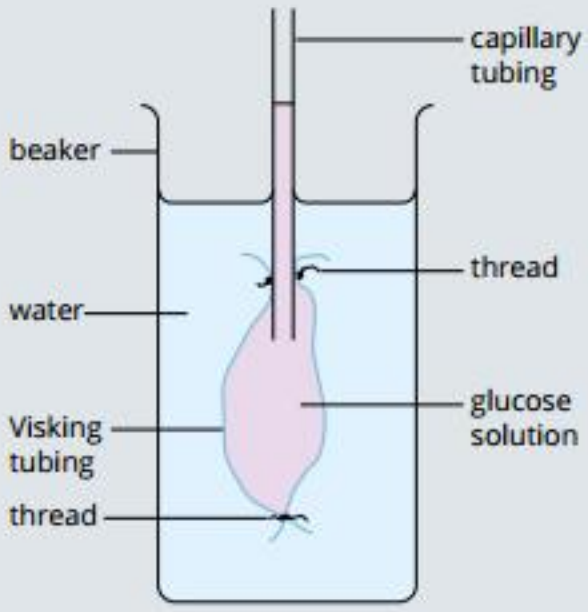


Analyse

22 Red blood cells have a nucleus when first formed, however, this is lost during the process of differentiation and maturation. The loss of the nucleus results in the shape of the cell changing from a sphere to a bi-concave disc. Explain how this change assists red blood cells to perform their function.

23 Compare active and passive transport.

24 Visking tubing is made of cellulose. It acts as a semipermeable membrane and can be used to model the cell membrane. Visking tubing has pores that range between 1 and 10 nm in diameter. Glucose molecules have a diameter of 0.7 nm and water is 0.00031 nm. An experiment was undertaken by a group of students to investigate the effect of solute concentration on the diffusion of glucose molecules across the Visking tubing. The set up used is shown below:

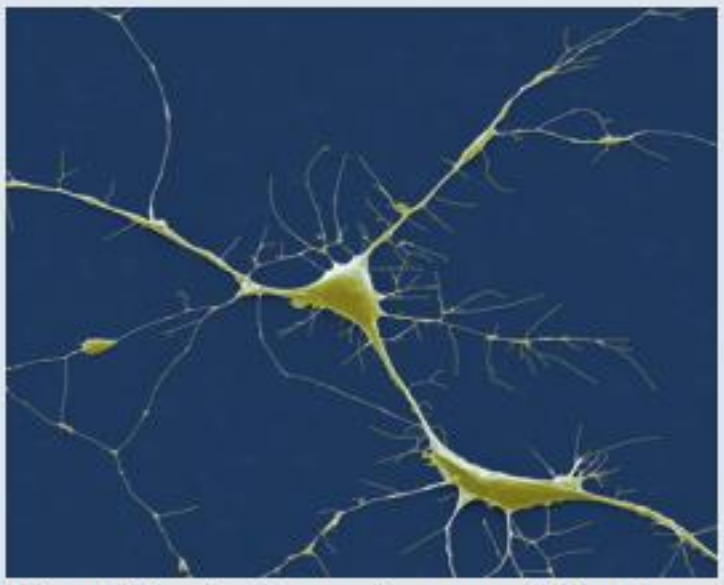


Students calculated the change in volume over a lesson at three different glucose concentrations (0.5 M, 1.0 M and 2.0 M). The class average results are shown in the table below.

Time (m)	% change in volume 0.5 M	% change in volume 1.0 M	% change in volume 2.0 M
0–3	1.5	2.8	4.4
3–6	1.1	2.2	3.3
6–9	0.9	2.1	3.0
9–12	0.9	2.0	2.9
12–15	0.8	1.9	2.9
15–18	0.8	1.7	2.6
18–21	0.8	1.7	2.5
21–24	0.8	1.7	2.4
24–27	0.7	1.6	2.3
27–30	0.6	1.6	2.3

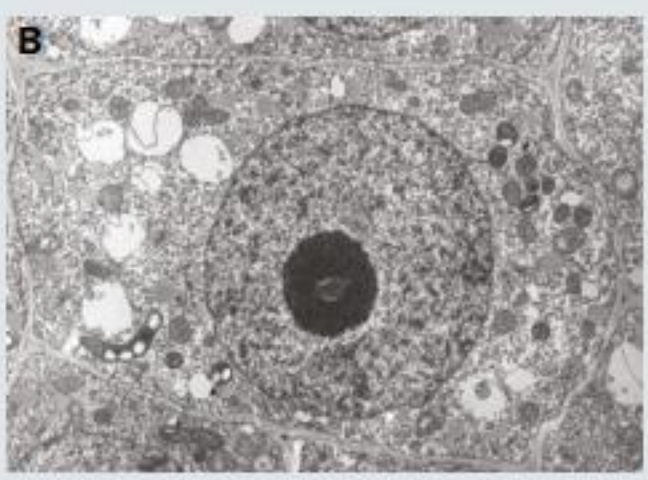
- **a** During which period of the experiment was the rate of osmosis the fastest? Justify your answer.
- **b** This experiment ran over 30 minutes, predict what would be seen if the experiment was left for 24 hours. Explain what would occur.

25 The following image is a nerve cell. With specific reference to the visible structures of the cell, deduce whether this image was taken using an electron microscope or a light microscope.



26 Visking tubing is often used as a model for the cell membrane. Discuss how accurately it truly represents the cell membrane.

27 Below are two cells observed under a scanning and a transmission electron microscope.



a One of the two cells is from a prokaryote. Explain which one.

b Determine if the eukaryotic cell is from an animal or a plant.

28 You are given a microscope slide with a sample of cells smeared on it and asked to identify the cell type. The cells are circular with a dark round mass at their centre. You estimate that the cells are approximately 20 µm in diameter.

a Classify the cells as prokaryotic or eukaryotic cells.

b Infer what organelle the dark round mass at the centre of the cells could be.

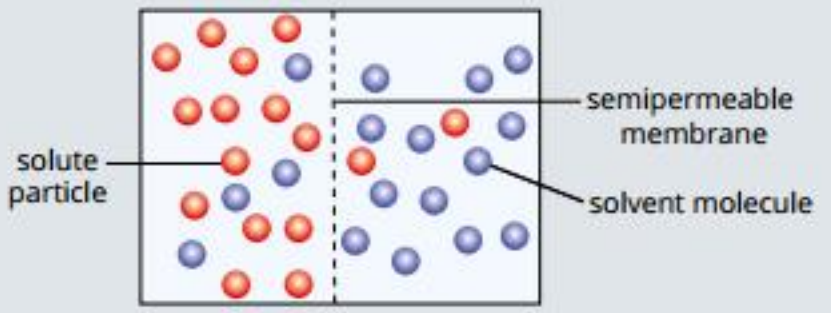
29 Using your knowledge of cells and surface area-to-volume ratio, infer the most effective body shape and surface area-to-volume ratio of an organism to:

a gain heat from its environment

b prevent heat loss

c maximise heat loss.

30 Two solutions are separated by a semipermeable membrane as illustrated below.



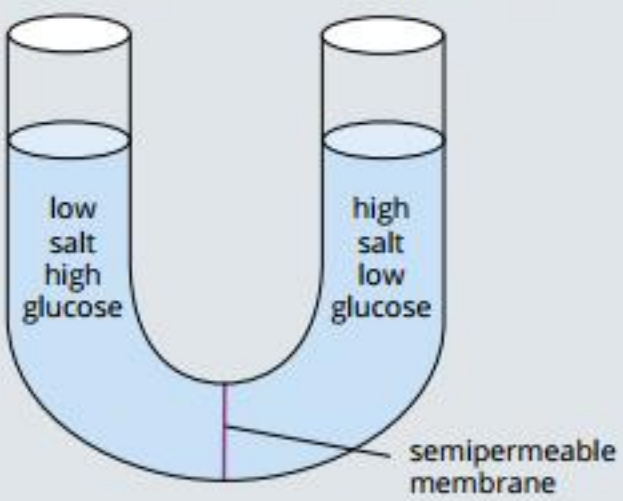
In which direction (if any) there would be a net movement of particles.

31 Two different solutions with the same volume are placed on either side of a semipermeable membrane in a U-shaped glass tube, as shown in the following diagram. The membrane is permeable to salt but not glucose. The tube is then left to stand for several days. Predict what would happen to the:

a salt concentration on each side of the membrane

b glucose concentration on each side of the membrane

c fluid levels on each side of the membrane.



32 In mammals, cells lining the:

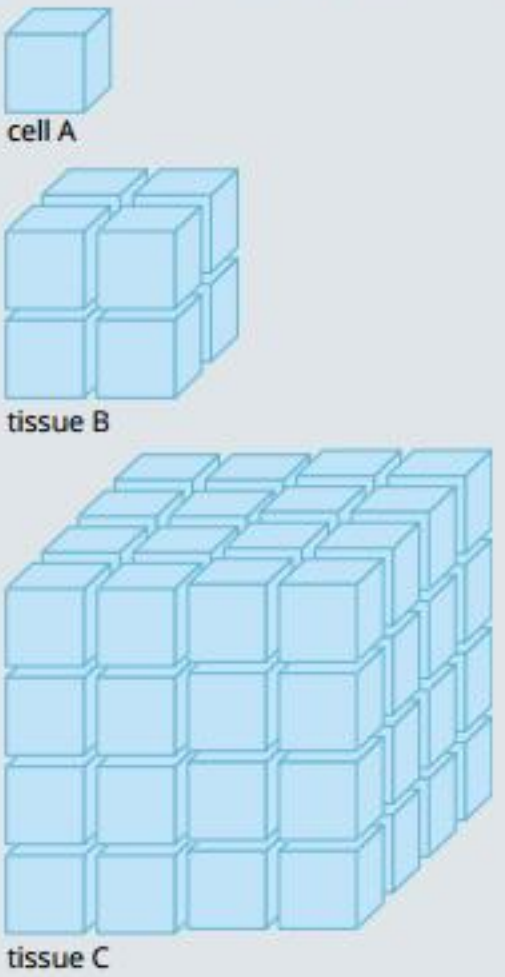
a alveoli of the lung take up oxygen by diffusion

b tubules of the kidney take up glucose by active transport

c small intestine take up fat droplets by pinocytosis.

Explain why the different methods of uptake are appropriate for the substances taken up in each case.

33 The following diagram represents living cells. Cell A, tissue B and tissue C all have the same volume.



Determine which one of the follow statements is correct.

A In distilled water, tissue B would gain water at a greater rate than cell A.

B In distilled water, the cells in tissue C would shrink at a greater rate than cell A.

C In a concentrated salt solution, tissue C would gain water at a greater rate than cell A.

D In a concentrated salt solution, tissue C would lose water more slowly than tissue B.

Interpret

34 It has been hypothesised that animals living in extreme temperatures will contain higher levels of cholesterol in their cell membranes in order to stabilise the membrane's structure. An investigation was undertaken into this idea. The membranes of liver cells from four species of fish were analysed to determine the amount of cholesterol present. Three species of Antarctic fish and one from temperate waters were used. The data collected is shown below.

Species	Habitat	Cholesterol (µmol/g of tissue)
Trematomus bernacchii	Antarctic	0.155
Pagothenia borchgrevinki		0.125
Chionodraco hamatus		0.11
Notolabrus celidotus	Temperate	0.17

Does the presented data support or disprove the hypothesis? Justify your answer.

35 A new unicellular organism has been discovered by light microscopy. Its characteristics include:

- internal membrane-bound circular structures composed of DNA
- two whip-like structures located close to each other at one end of the cell
- a semirigid structure outside the cell membrane
- a length greater than its width
- a chloroplast.

In your studies on cell biology, you have identified six main groups of organisms based on their cell structures: plant cells, animal cells, fungal cells, protists, bacteria and archaea.

Hypothesise which group this new organism would most likely belong to and give two reasons to support your answer.

36 At the cellular level, materials move through the cell membrane by several processes. At the organ level, the exchange of materials is facilitated by the arrangement of cells, which provides a large surface area. Discuss this statement with specific reference to the processes by which materials move through the cell membrane. Outline three of the processes. For each process:

a give an example of a material taken up

b state where this uptake occurs

c explain why the process is appropriate.

Data analysis

DATA SET 1

The information below applies to Questions 1–5.

An experiment was conducted to investigate the effects of increased CO_2 levels on the mitochondria of a number of plant species. Plants were either grown in normal room air or in an environment with increased CO_2 levels. The number of mitochondria per 100 μm^2 of cell area was determined. The size of the mitochondria in all plants were equivalent in both normal and increased CO_2 conditions.

The data gathered from the experiment is shown in Table 1.

TABLE 1 The number of mitochondria in normal CO_2 and increased CO_2 conditions in different plant species.

Species	Mitochondria per 100 μm^{-2} cell area		Ratio increased/normal
	Normal CO_2	Increased CO_2	
Glycine max	1.0 ± 0.1	1.6 ± 0.1	
Abutilon theophrasti	0.3 ± 0.03	0.4 ± 0.03	
Pinus radiata	14.0 ± 1.4	30.0 ± 1.8	
Piper auritum	0.4 ± 0.05	0.9 ± 0.08	
Pinus taeda	0.5 ± 0.07	0.9 ± 0.10	
Acer rubrum	0.3 ± 0.03	0.6 ± 0.06	
Liquidambar styraciflua	0.7 ± 0.06	1.5 ± 0.22	
Cercis canadensis	1.3 ± 0.15	2.4 ± 0.22	

Question 1 (4 marks)

Calculate the mean number of mitochondria per 100 μm^{-2} cell area for both normal and raised CO_2 levels.

Question 2 (2 marks)

Identify any outliers.

Question 3 (2 marks)

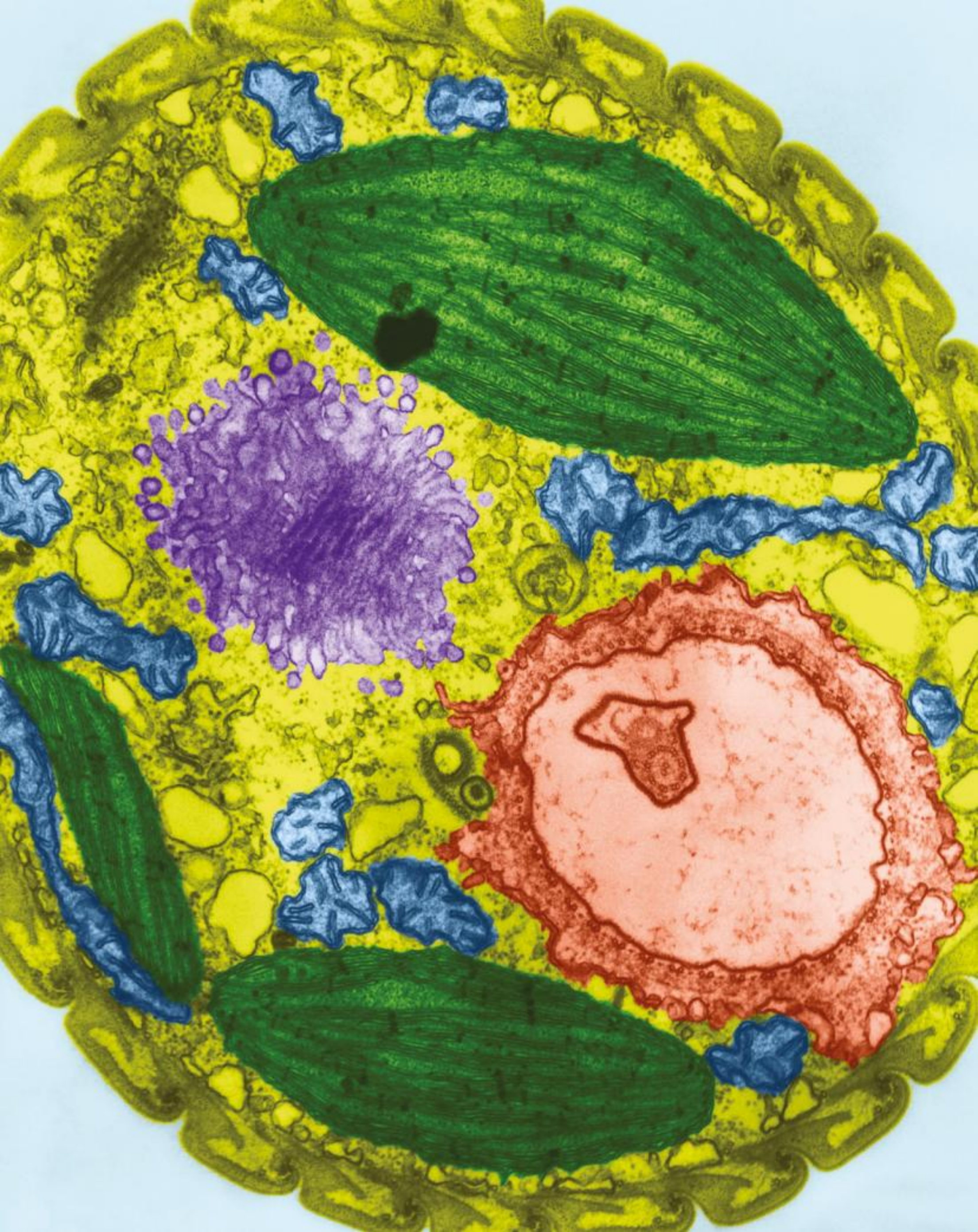
Complete the table by calculating the ratio of increased/normal mitochondria 100μm^{-2} cell area.

Question 4 (2 marks)

Discuss any trends in the data.

Question 5 (1 mark)

What conclusions, if any, can be drawn about the effects of increased CO_2 levels on mitochondrial numbers in plants?



CHAPTER

03 The functioning cell

Because cells are the smallest discrete living unit, they are acknowledged as the building blocks of living organisms. It was only in the 20th century, when the fields of enzymatics and proteomics were developed, that scientists came to really appreciate cellular organelles and the dynamic biochemical world that exists within a single cell. Every cellular function and each step of every metabolic pathway, depends on the actions of a group of molecules called enzymes.

In this chapter, you will learn about the key types of biomolecules. You will come to appreciate the complex structure of enzyme molecules, as well as factors that influence their ability to function. As you journey down the metabolic pathways of photosynthesis and cellular respiration, you will come to value their importance in the life of every living organism on this planet.

Syllabus subject matter

Topic 2 • Exchange of nutrients and wastes

- Describe the structure and function of carbohydrates, proteins and lipids.* **3.1**
- Describe the structure and function of enzymes, including the role of the active site. **3.2**
- Compare the induced-fit and lock-and-key models of enzyme function. **3.2**
- Explain how enzyme activity is affected by factors such as temperature, pH, presence of inhibitors and substrate concentration. **3.2**
- Interpret data from an experiment investigating factors affecting enzyme activity. **3.2, Data analysis, Practical investigation 3**

Topic 3 • Cellular energy, gas exchange and plant physiology

- Distinguish between catabolism and anabolism. **3.2**
- Explain how ATP allows energy from catabolic reactions to be used in anabolic reactions. **3.3**
- Describe the process of aerobic respiration, identifying the location in the cell and net inputs and outputs of:
  - glycolysis
  - Krebs cycle and electron transfer chain
  - the overall reaction ($C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O$ + 36–38 ATP). **3.3**
- Compare aerobic and anaerobic respiration. **3.3**
- Describe the process of photosynthesis, identifying the location in the cell and net inputs and outputs of:
  - light-dependent reactions
  - light-independent reactions
  - the overall reaction ($6CO_2 + 6H_2O$ + light energy $\rightarrow C_6H_{12}O_6 + 6O_2$). **3.4**

* The greyed-out dot point is also addressed in Chapter 5.

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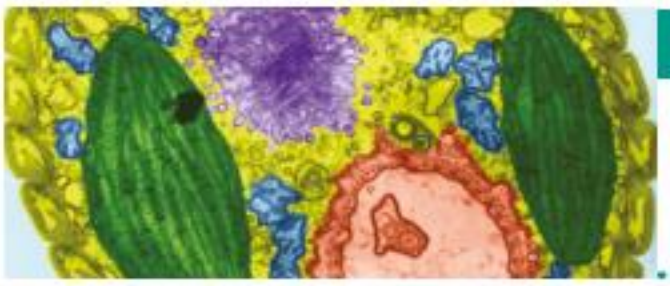
3.1 Molecular composition of organisms



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- distinguish between organic and inorganic molecules
- identify the four main types of organic molecules
- understand the biological role of each type of organic molecule.

All life is composed of the same few chemical elements. There are 92 naturally occurring elements. Only 11 of these are found in organisms in more than trace amounts, and four of these—carbon (C), hydrogen (H), oxygen (O) and nitrogen (N)—make up 99% of organisms by mass. The same elements are also found in rocks, soil and air. However, there is a difference in the way that these atoms are organised into the huge variety of larger compounds in living organisms. Organisms, such as the plants and animals in Figure 3.1.1, produce compounds containing carbon and hydrogen, that are known as organic compounds. All other compounds, whether in living or non-living things, are called inorganic compounds.

An element is a substance that contains one or more atoms that have the same number of protons.

A molecule contains two or more atoms chemically joined together.

A compound is a molecule that contains at least two different elements.

FIGURE 3.1.1 A river in the Daintree rainforest in North Queensland. The plants, animals, rocks and water found in the rainforest all contain molecules. Only the plants and animals produce organic compounds.

In this module, you will learn about the difference between organic and inorganic compounds. In addition, you will explore the four main types of organic molecules—nucleic acids, carbohydrates, lipids and proteins.

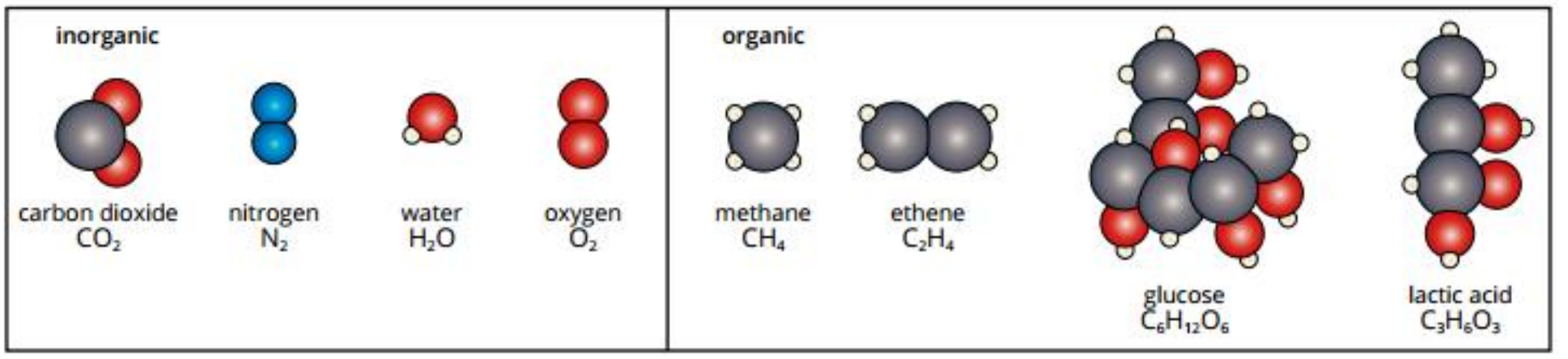


FIGURE 3.1.2 Models of some common molecules in organisms. Carbon atoms are coloured black, oxygen are red, hydrogen are white and nitrogen are blue.

ORGANIC MOLECULES

Organisms produce complex compounds that contain carbon and hydrogen, as shown in Figure 3.1.2 (on page 58). These are called organic compounds because the first compounds discovered were either produced by or found in organisms. Most large organic molecules are built from many smaller units of organic molecules linked together, e.g. proteins are built from amino acids.

All other elements and compounds, whether in living or non-living things, are referred to as inorganic. Inorganic substances that are important for living organisms include water **(H_2O)**, **oxygen (O_2)**, **carbon dioxide (CO_2)**, nitrogen (N) and minerals like magnesium (Mg^{2+}), iron (Fe^{2+}) or calcium (Ca^{2+}).

The four main types of organic molecules are **carbohydrates**, proteins, nucleic acids and lipids. Carbohydrates, proteins and nucleic acids are large chainlike molecules called polymers (*polys*, meaning 'many', and *meros*, meaning 'part'). Polymers are formed by joining together many smaller units (monomers) to form a chain.

In organisms, organic molecules can be converted from one form into another. Units may be linked together to form larger molecules. For example, **glucose** units may be linked together to form larger carbohydrates such as cellulose, shown in Figure 3.1.3. Other chemical groups may be attached to form molecules such as glycoproteins (proteins with sugars attached, Figure 3.1.4) and phospholipids (lipids with phosphate attached, such as in the cell membranes studied in Module 2.3 and seen in Figure 2.3.3). When food is plentiful, carbohydrates are converted into fats for storage. When food is scarce, the reverse occurs and even proteins can be converted into small molecules to use for energy.

Any molecule that is found in a living organism is called a biomolecule. Examples of biomolecules are fatty acids, carbohydrates and hormones. Many large biomolecules are called biomacromolecules, and many are polymers. Polymers can be made up of thousands of atoms and include proteins and nucleic acids. Polymers are built of smaller repeating units called monomers.

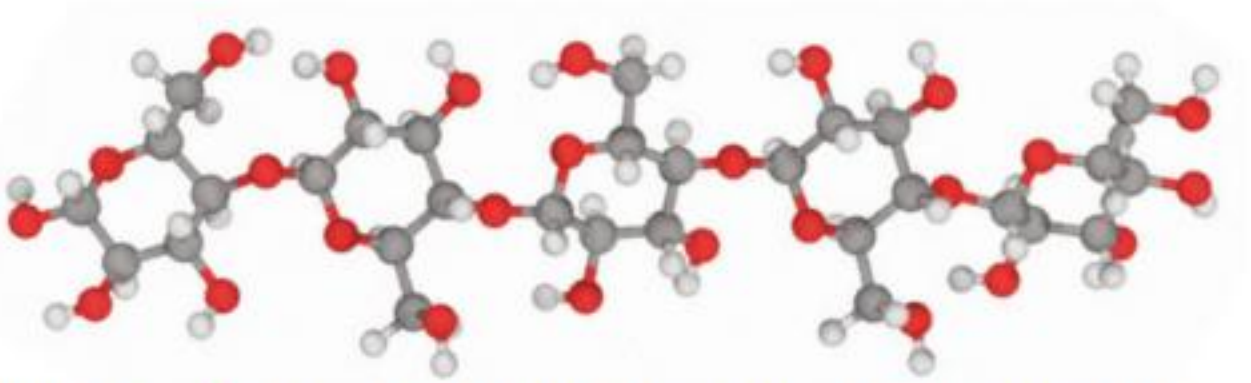


FIGURE 3.1.3 Cellulose is a polymer. A strand of cellulose is made of several glucose molecules joined together. Each ring in this molecular model represents a glucose.

FIGURE 3.1.4 A computer model of a fibrillin glycoprotein. The protein component is represented by ribbons and the sugars are represented by the ring structures.

Carbohydrates are organic compounds, such as sugars, starch and cellulose, that are made of carbon, hydrogen and oxygen. Carbohydrates as a class include monosaccharides, disaccharides and polysaccharides (the complex carbohydrates) as seen in Figure 3.1.5. Only polysaccharides are polymers.

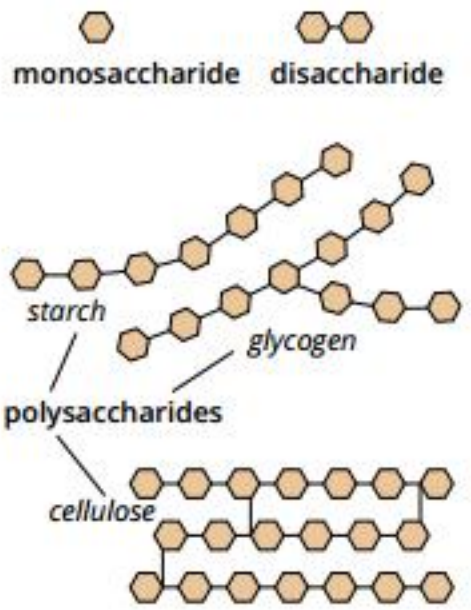


FIGURE 3.1.5 The structures of some carbohydrates showing how they are built from subunits

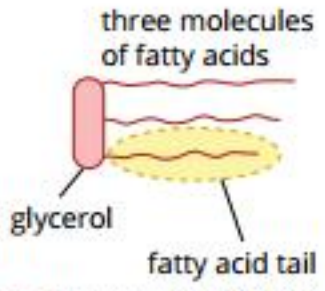


FIGURE 3.1.6 The structure of lipids

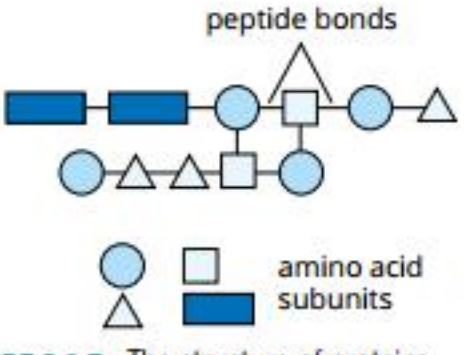


FIGURE 3.1.7 The structure of proteins

Carbohydrates

Carbohydrates are the most abundant organic molecules in nature. They are an important source of chemical energy for living organisms (e.g. glucose). Carbohydrates are also used for energy storage; in plants, energy is stored as starch, while in animals, energy is stored as glycogen. Carbohydrates, in the form of cellulose, are also used for structural support in plants (Figure 3.1.5).

Carbohydrates are compounds made of carbon, hydrogen and oxygen. There are three main groups of carbohydrates: monosaccharides, disaccharides and polysaccharides. You can see these in Figure 3.1.5. Examples of monosaccharides include glucose, fructose and galactose. Although these three monosaccharides have the same chemical formula ($C_6H_{12}O_6$), each molecule has a different structure, and therefore has different properties.

When two monosaccharides are joined together, they form a disaccharide (meaning 'two sugars'), and a molecule of water is removed. For example, milk sugar (lactose) is made from glucose and galactose, and cane sugar (sucrose) is made from glucose and fructose. The two disaccharides have the same chemical formula ($C_{12}H_{22}O_{11}$), but the atoms are arranged differently, causing the molecules to have different properties. When many sugars are joined together, they form biomacromolecules called polysaccharides ('many sugars'). Some polysaccharides are composed of one type of monomer, such as starch and cellulose. Complex polysaccharides consist of different monosaccharide subunits in the same molecule, such as murein found in the cell walls of bacteria.

Lipids

Lipids are fatty substances that consist of non-polar hydrophobic (resistant to water) molecules. They include fats and oils, which are important as energy-storing molecules. Figure 3.1.6 shows the structure of some common lipids called triglycerides but not all lipids are built the same way. Lipids are not classified as true polymers because they are not all made from the same repeating units. Phospholipids are an important component of cell membranes, and they have two fatty acid tails. These membranes enclose the cell's contents and subdivide it into the many sub-cellular organelles. Steroids are a group of lipids that act as membrane components, hormones and vitamins.

Lipids are composed of carbon, hydrogen and oxygen, but in different proportions to carbohydrates. Lipids contain a much smaller proportion of oxygen and often contain hydrocarbon chains. Lipids can also contain other elements such as phosphorus and nitrogen.

Proteins

Proteins are more complex than carbohydrates or lipids. There are thousands of different kinds of proteins, and their functions vary widely. While carbohydrates and lipids are similar in all plants and animals, each kind of organism has its own unique proteins. Some proteins form structural components of cells; others are **enzymes**, hormones or carrier molecules. For example, haemoglobin is a protein that carries oxygen in the blood. To form enough functional haemoglobin for production of new red blood cells, bone marrow needs a good supply of the mineral iron (Fe) as seen in Figure: 3.1.8. All proteins contain carbon, hydrogen, oxygen and nitrogen; many also contain sulfur and often phosphorus and other elements. Proteins are polymers composed of chains of smaller subunits (monomers) called **amino acids**, as shown in Figure 3.1.7. Amino acids in proteins are linked by a particular kind of chemical bond called a peptide bond, and proteins are formed from polypeptides or polypeptide chains (Figure 3.1.7). There are 20 different amino acids commonly found in human proteins.

The study of all the proteins of an organism is known as **proteomics**. In medicine, 99% of all drugs are proteins, or act by binding to proteins. A better understanding of proteins and proteomes (all the proteins produced by an organism) will help develop new pharmaceuticals, clarify the relationships between genes and diseases (e.g. by identifying marker proteins for diseases) and lead to better treatments. The complexity and importance of proteomics has led to the establishment of an international Protein Data Bank (PDB). The PDB can be freely accessed online by searching for PDBsum. It holds a pictorial database of many hundreds of thousands of models of protein structures, such as the ribbon diagram of haemoglobin in Figure 3.1.8.

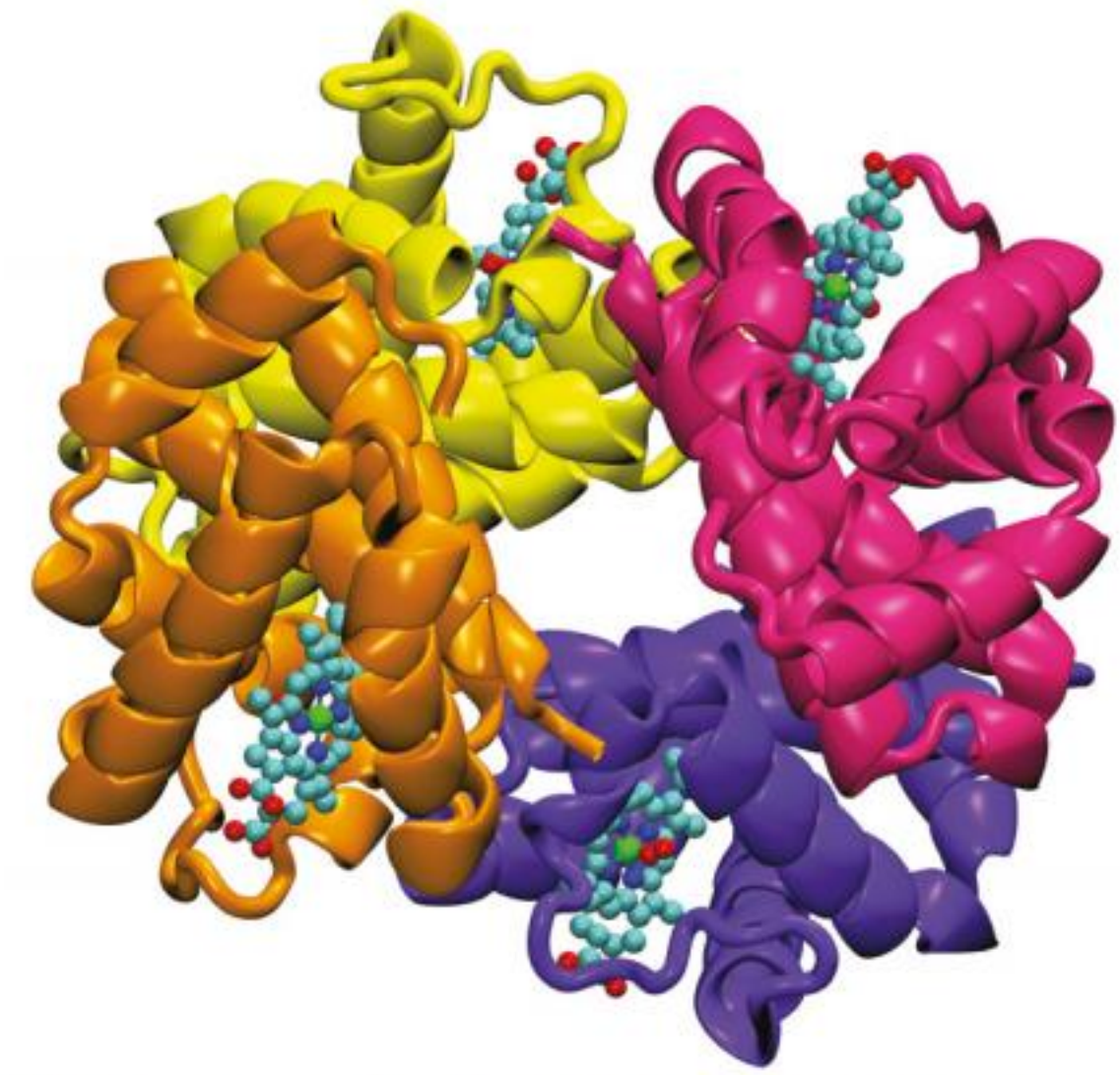


FIGURE 3.1.8 Haemoglobin is made up of four protein subunits (coloured ribbon structures). Each subunit has an oxygen-binding site called the haem group (turquoise). Within each haem group there is one atom of iron (green). Oxygen molecules are shown as paired red spheres.

Nucleic acids

Nucleic acids are the genetic material of all organisms, and they determine many of the features of an organism. Nucleic acids are polymers composed of long chains of subunits called nucleotides. A nucleotide consists of a phosphate, a sugar and a nitrogenous base, as shown in Figure 3.1.9. You will learn more about the nucleic acids later.

There are two types of nucleic acids:

- deoxyribonucleic acid (DNA), which carries the 'instructions' for assembling proteins from amino acid subunits using a genetic code. DNA is passed accurately from cell to cell during cell division. The four nitrogenous bases in DNA are adenine (A), thymine (T), guanine (G) and cytosine (C)
- ribonucleic acid (RNA), which plays a major role in the manufacture of proteins within cells. The four bases in RNA are adenine (A), uracil (U), guanine (G) and cytosine (C).

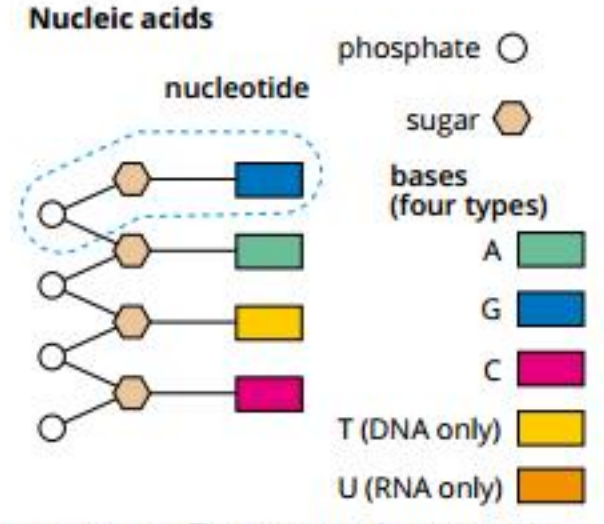


FIGURE 3.1.9 The structure of nucleic acids

FIGURE 3.1.10 Mountain ash trees (*Eucalyptus regnans*) are the tallest of all flowering trees and can reach a height of up to 114 m. Cohesion between water molecules holds the water together and allows the water to be drawn up the trunks of the trees.

Vitamins

Vitamins are organic molecules that animals require in small amounts for normal functioning. Animals can synthesise some vitamins but must obtain others in their diet. For example, humans synthesise Vitamin D in skin cells exposed to sunlight, and while most mammals can synthesise vitamin C, humans must obtain it from their diet. Vitamins may be water-soluble (such as vitamins B and C) or lipid-soluble (such as vitamins A, D, E and K). Water-soluble vitamins must be consumed regularly in the diet because they cannot be stored in body tissues. Lipid-soluble vitamins can be stored. Many vitamins have an important role to activate specific enzymes and are known as cofactors or coenzymes.

INORGANIC MOLECULES

Water

Life evolved in water. Most organisms are 70–90% water, and the chemical reactions in cells take place in a watery medium. This is why the properties of water, such as neutral pH, cohesiveness and heat capacity, are important in many biological processes. The neutral pH (7.0) of the large water volume helps to buffer against pH changes that may affect biochemical reactions. Many essential chemical reactions have water as a reactant or product, e.g. photosynthesis and cellular respiration.

Water molecules are very **cohesive**, meaning they have a strong tendency to stick together. This property allows thin columns of water to be pulled up tree trunks without breaking (Figure 3.1.10). Cohesive bonds between surface molecules of water lead to surface tension, which allows small insects, such as the pond skater in Figure 3.1.11, to walk across the surface of water without sinking.

Water has a high heat capacity—it can absorb a great deal of heat with very little increase in temperature. This is important for temperature regulation in organisms. When you exercise, the chemical reactions taking place in your cells produce heat. Much of this heat can be absorbed by the water in your body, without the cells heating up significantly. Because liquid water draws in a high amount of heat energy when it converts to vapour, the evaporation of even small amounts of water is effective in cooling that part of the body surface (Figure 3.1.12).

FIGURE 3.1.11 A common pond skater (*Gerris lacustris*) can walk on water. Notice that the legs of the pond skater seem to 'press' on the surface of the water.

FIGURE 3.1.12 A coloured scanning electron micrograph of the skin surface of the back of a human hand, showing sweat droplets (blue). Evaporation of water on the skin surface is a method of controlling body temperature.

Oxygen and carbon dioxide

Most cells need oxygen (O_2) to release energy from food molecules in processes known collectively as **cellular respiration**. Therefore, a constant supply of oxygen is necessary to maintain the activity of these cells. It is usually easy for organisms that get their oxygen from air because the atmosphere is 21% oxygen by volume (Figure 3.1.13). However, oxygen is not very soluble in water, so organisms that get their oxygen from water either are small, flat and relatively inactive, or have very efficient ventilation systems with a large surface area for gaseous exchange, such as the fish gills in Figure 3.1.14.

Carbon is the key atom in organic molecules, with many organic molecules being formed initially from the CO_2 in the atmosphere. Carbon dioxide (CO_2) is taken from the atmosphere (which has only 0.035% by volume of carbon dioxide, Figure 3.1.15) by plants, some bacteria and some protists. It is used in the process of **photosynthesis** to make sugars. These sugars are the start of a food chain when the autotrophs (usually plants) are eaten by the heterotrophic animals. Carbon dioxide is eventually returned to the atmosphere, mainly by the decay of organic material and as an end product of cellular respiration. This cycling of carbon through organisms and the atmosphere is critical to the survival of all organisms.

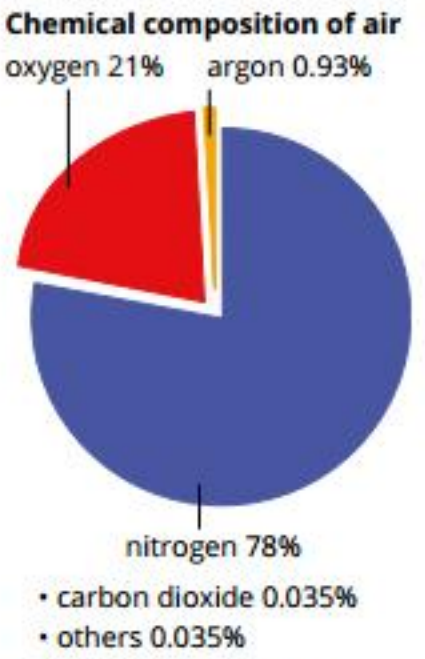


FIGURE 3.1.13 Air is a mixture of mainly nitrogen, oxygen and very small amounts of other gases. Carbon dioxide, used in photosynthesis by plants, is only 0.035% by volume under natural conditions.

Nitrogen

Organisms require nitrogen in relatively large amounts because it is a key component of all proteins, nucleic acids and the ATP energy-carrier molecule. Potentially, there is an abundance of nitrogen available because the atmosphere has approximately 78% nitrogen gas (Figure 3.1.13), however, most organisms cannot use nitrogen in its gaseous form. Some bacteria and cyanobacteria convert atmospheric nitrogen into compounds that can be used by plants such as ammonia [NH_3] and soluble nitrate ions [NO_3^-]). This process is known as nitrogen fixation. The most important nitrogen-fixing bacteria are the symbiotic bacteria found in nodules in the roots of plants, including legumes (peas and beans), casuarinas and acacias (Figure 3.1.15).

Nitrogen compounds produced by the soil bacteria are absorbed by plants and used to make amino acids. Heterotrophs obtain their amino acids (needed to make proteins) in food chains by consuming plants and other organisms. They also produce nitrogen-rich waste (manure), which has traditionally been used as a plant fertiliser.

FIGURE 3.1.14 A coloured scanning electron micrograph of mackerel (*Scomber scombrus*) gills, showing the large surface area for gaseous exchange.

FIGURE 3.1.15 (a) Nodules containing nitrogen-fixing bacteria on the roots of a garden pea (*Pisum sativum*). (b) A coloured scanning electron micrograph of nitrogen-fixing soil bacteria (*Rhizobium* species) in a root nodule of a bean plant. These bacteria (green) have a symbiotic relationship with the plant.

FIGURE 3.1.16 A soil profile showing the horizons (layers), which vary in colour depending on the mineral content in the soil. Plants absorb these minerals when they draw water out of the soil.

FIGURE 3.1.17 Bone matrix is made up of inorganic components including salts of calcium and phosphorus.

Minerals

Mineral salts are naturally occurring inorganic compounds produced by weathering of rocks. The water-soluble mineral salts are then absorbed as ions into the roots of plants (Figure 3.1.16), making them available to be eaten by animals. Humans require more than 20 minerals. Biologically important minerals include phosphorus, potassium, calcium, magnesium, iron, sodium, iodine and sulfur. Many others are needed in small (trace) amounts.

Mineral ions are found in the cytosol of cells, in structural components (such as bone), and in the molecules of many enzymes and vitamins. They may also be incorporated into other important organic compounds in cells. Examples are shown in Table 3.1.1.

TABLE 3.1.1 Minerals: their functions and where they are found

Mineral	Function	Present in
phosphorus (P)	part of ATP (main energy carrier molecule) in cell membranes	• animals • plants
magnesium (Mg)	• part of chlorophyll • muscle and nerve function; bone development • molecule used for photosynthesis	• animals • plants
iron (Fe)	• part of haemoglobin molecule and many metabolic functions • chloroplast function and function • chlorophyll synthesis	• form haem in animals (Figure 3.1.8) • non-haem in plants • haem in plants
calcium (Ca)	• bone and teeth structure, muscle function • root growth	• animals (Figure 3.1.17) • plants
potassium (K)	• muscle and nerve function • transport functions	• animals • plants
sodium (Na)	• muscle and nerve function • controls osmotic pressure in cells	• animals • plants

3.1 Review

SUMMARY

- Organic components include carbohydrates, lipids, proteins and nucleic acids. All except lipids are polymers.
- Inorganic components of living organisms include water, oxygen, carbon dioxide, nitrogen and minerals such as calcium.
- Carbohydrates are important as energy sources and for structural components. They are similar in most organisms.
- Lipids play an important role in cell membranes and are similar in most organisms.
- Proteins are composed of amino acids and have many vital functions. Each type of organism has their own unique group of proteins.
- Nucleic acids carry the genetic information of cells and are built from chains of subunits called nucleotides.
- Vitamins are smaller organic molecules that are vital for normal cell function.
- Minerals are inorganic chemicals important in limited but necessary amounts for body structure and function.
- Important properties of water include cohesiveness, surface tension, heat capacity and a neutral pH.
- Oxygen is needed for efficient energy supply in cells.
- Carbon dioxide is the original source of carbon for organic molecules.
- Nitrogen is a key atom in protein, nucleic acid and ATP molecules.

KEY QUESTIONS

Describe

1 Use examples to demonstrate the difference between an 'organic compound' and an 'inorganic compound'.

2 Define 'polymer' and 'monomer'.

3 Create a table to list the three groups of carbohydrates with an example for each.

4 Identify the atom that makes proteins different from carbohydrates and lipids.

5 What are the monomer units of polypeptides?

6 Describe the four biomolecules. Indicate whether or not they are polymers, and describe the subunits involved.

Apply

7 Explain the structure of proteins.

8 Determine whether carbon dioxide is organic or inorganic. Explain why.

9 **a** Explain the importance of nitrogen for living organisms.

b Summarise what nitrogen-fixing bacteria are and why they are important.

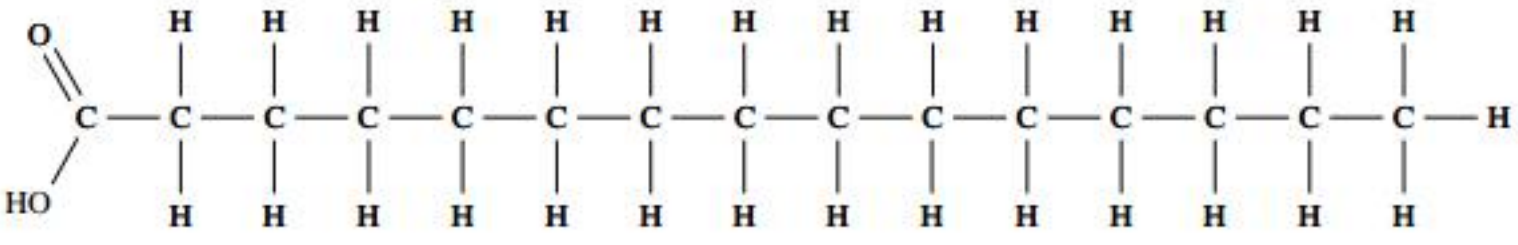
10 Distinguish between mono-, di- and polysaccharides.

11 Water is sometimes called the 'universal solvent'. Explain how this property is valuable to living things.

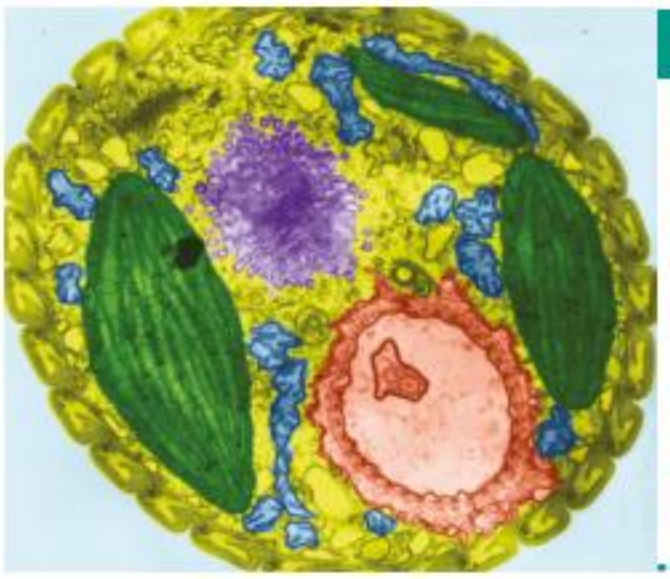
12 Apply your understanding of the human body to recognise five examples of how proteins are vital to body function.

Analyse

13 Determine the type of biomolecule represented by the image below.



3.2 Enzymes and biochemical pathways



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that enzymes act as biological catalysts to initiate chemical reactions
- describe the structure and function of enzyme molecules
- understand the role of an enzyme's active site
- recognise that induced-fit is currently the accepted model for enzyme function and compare it with the lock-and-key model
- explain how an enzyme's rate of reaction is influenced by temperature, pH, inhibitors and substrate concentration
- distinguish between anabolism and catabolism in biochemical reactions.

Nearly every function of a living organism depends on protein molecules. The functions of proteins include speeding up biochemical reactions, playing a role in cellular communication, in movement, storage and even structural support. Keratin in human hair, nails and the outer skin layer is a protein. Proteins are an essential component of cell membranes and the membranes that enclose most organelles. A human has tens of thousands of different proteins. Each protein has a specific sequence of sub-units (monomers) called amino acids, giving it a unique shape that enables it to carry out its particular function.

In this module, you will learn about the features of an essential subset of proteins, the enzymes. You will learn about each enzyme's specificity for a particular substrate, and how they interact with substrates to catalyse biochemical reactions. You will also learn about the factors that may affect an enzyme's activity.

ENZYME STRUCTURE

As with every protein, enzymes are formed by linking together a sequence of amino acids. This sequence is controlled by the cell's DNA. The primary chain of amino acids is then meticulously folded into a secondary structure, and coiled into a tertiary structure forming a precise and functional, three-dimensional, globular shape. Figure 3.2.1 illustrates the folding of amino acid chains into a functional enzyme molecule.

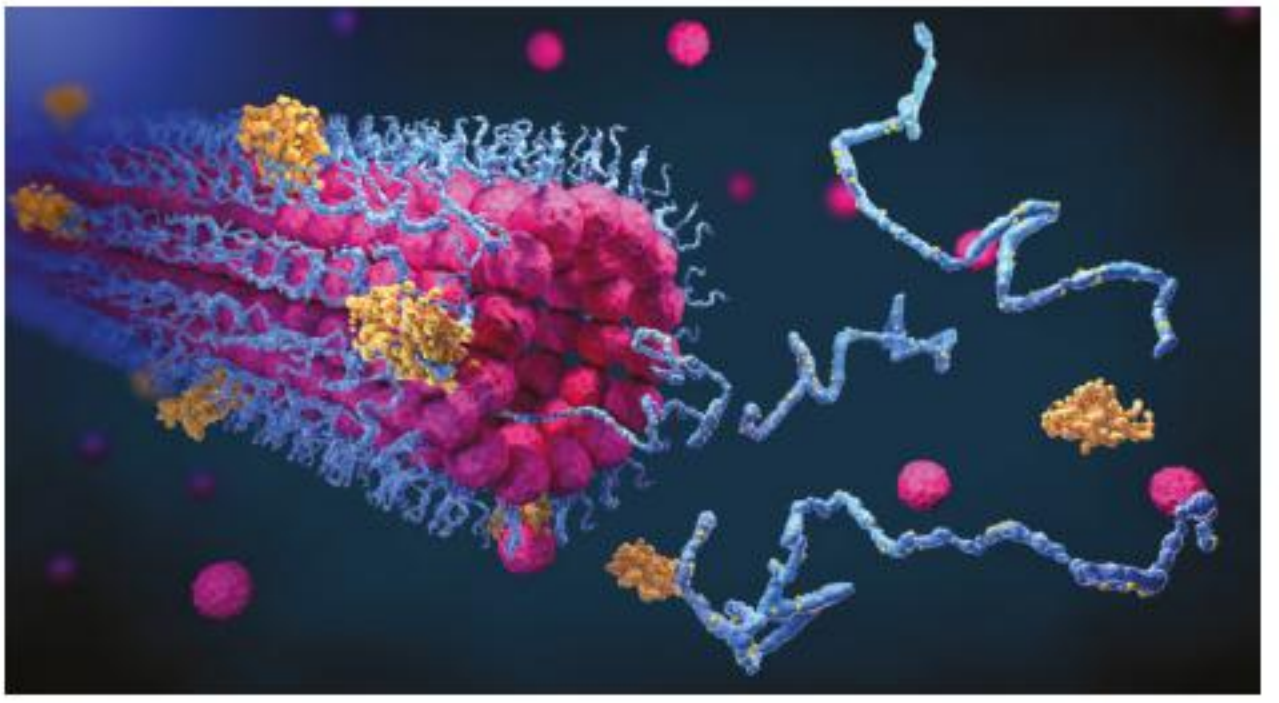


FIGURE 3.2.1 Enzymes are complex protein molecules that act as catalysts for biochemical reactions. They are formed from chains of many amino acids folded and coiled into specific three-dimensional structures.

Most enzymes are complex globular proteins with one or more grooves around their surface. These grooves are called **active sites** and are fundamental to enzyme function. Active sites are unique in their size and shape, and only specific types of other molecules will fit into them. The molecules that fit into an enzyme's active site are known as substrates. Most enzymes are highly specific and act only on a single **substrate**, although some enzymes can act on a group of similar substrates and therefore regulate many biochemical reactions.

Enzymes are often named after the substrate that they act upon, and usually have an *-ase* ending. For example, the enzyme, sucrase, breaks down sucrose sugar into its basic monosaccharides—glucose and fructose. Glucose and fructose can then be easily absorbed into the bloodstream and transported to your body cells where they are used as an energy source (Figure 3.2.2).

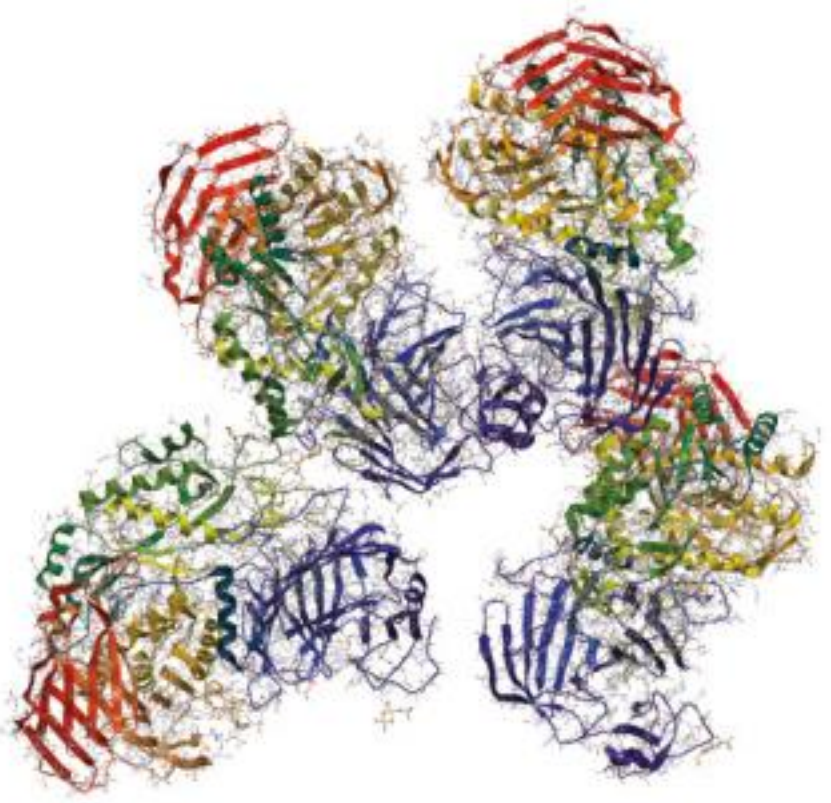


FIGURE 3.2.2 The enzyme sucrase (shown here as a computer-generated model) is located in the villi lining of your small intestine. It breaks down sucrose (a disaccharide) into simple monosaccharides, which are then absorbed into your bloodstream for transportation to cells.

ENZYME FUNCTION

Enzymes can join smaller molecules together to form a larger molecule. This occurs during an **anabolic reaction** (or anabolism). Other enzymes break down larger molecules into smaller ones, such as the sucrase-sucrose example above. This occurs during a **catabolic reaction** (or catabolism). In both cases, when the active site of an enzyme binds to a substrate, an **enzyme–substrate complex** is formed. Unlike the substrate, the specific enzyme is not changed during the reaction. It may be used repeatedly while that reaction is required and there is a substrate present in the cell. Enzyme reactions are represented for anabolism and catabolism in Figure 3.2.3.

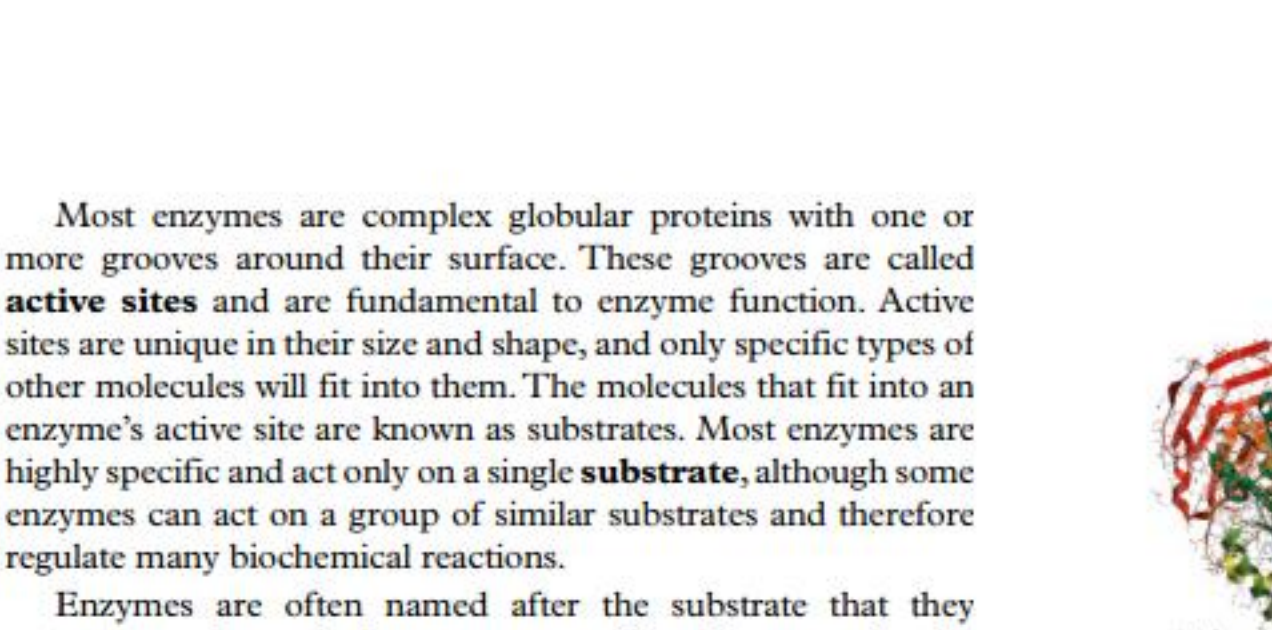


FIGURE 3.2.3 Enzyme reactions can be catabolic (enzyme A is breaking the substrate apart) or anabolic (enzyme B is joining molecules together).

Anabolic chemical reactions require an input of energy to build larger compounds from smaller molecules.

Catabolic reactions release energy as complex larger molecules are broken down into smaller ones.

For this reason, enzymes are called substrate-specific **catalysts**. Enzymes are considered catalysts because they speed up the rate at which biochemical processes occur without being used up in the process. They are substrate-specific because they regulate a limited number of biochemical reactions. Enzymes may have a high specificity, with the ability to act as a catalyst with one substrate only, or a low specificity and act as a catalyst for a range of similarly shaped substrates.

Enzyme molecules are often embedded in membranes. The more enzymes present, the more chemical reactions can take place. Hence, many organelle membranes that are sites of metabolic activity are intricately folded. This increases the available surface area, and the number of enzymes, many times over. An example of this is the elaborately folded inner membranes of mitochondria known as cristae. They are studded with ubiquinone and cytochrome C enzymes and are a vital part of ATP synthesis in the cell (Figure 3.2.4). You will learn more about ATP synthesis in Module 3.3.

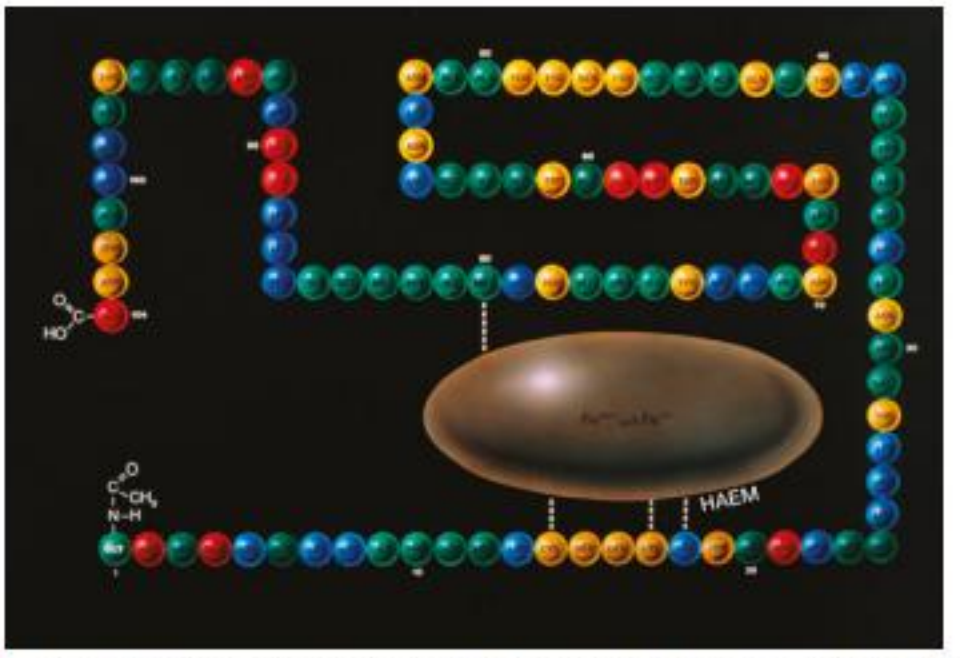


FIGURE 3.2.4 A model of the enzyme cytochrome C, important in the process of producing ATP for energy in a cell. Here, one cytochrome C molecule is seen. It is made of 104 amino acids with a 'haem' ring containing iron (Fe). Cytochromes are respiratory enzymes located in the membrane of organelles such as the mitochondria, chloroplasts and endoplasmic reticulum.

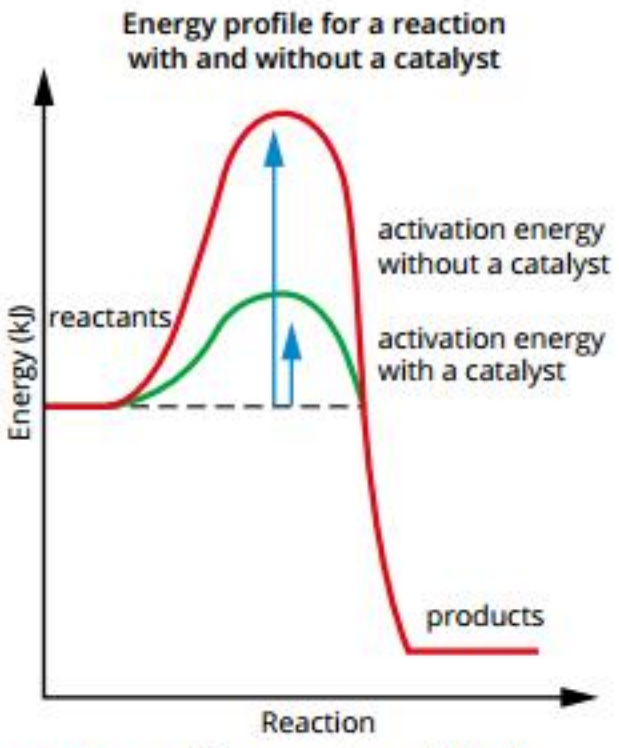


FIGURE 3.2.5 Enzymes act as catalysts to reduce the amount of energy required for substrate molecules to react with each other. This enables your cells to have millions of chemical reactions every minute at normal body temperature.

ENZYME CATALYSIS

At any given moment, there are thousands of biochemical reactions taking place within living cells. An enzyme's function is to control and regulate these biochemical processes. Energy is used to join molecules to form new larger compounds in anabolic reactions. Energy is released when larger compounds are broken apart to form new smaller compounds in catabolic reactions.

Every chemical reaction requires a level of **activation energy** to initiate the reaction. There may not be enough energy present for these reactions to take place at the rate required by cells without an enzyme catalyst. Enzymes operate by reducing the activation energy of biochemical reactions as shown in Figure 3.2.5

An enzyme orientates and holds substrate molecules in the correct position on the active site to reduce the activation energy required for changing chemical bonds in the substrate.

FACTORS AFFECTING ENZYME FUNCTION

The activity of enzymes can be affected by several factors, including **cofactors**, temperature, pH, inhibitors and the concentration of reactants and products.

Some enzymes will only be activated by the addition of a cofactor. Enzyme cofactors are either organic **coenzyme** molecules or inorganic ions, which are required for enzyme activity. Some cofactors are permanently attached to enzymes. Other cofactors only attach to an enzyme in response to a specific event in your body.

Temperature

High temperatures can directly affect the weak hydrogen bonds holding the enzyme into its specific molecular shape. Heat energy causes bonds to weaken, distorting the groove shape of the active site on the enzyme surface, thus preventing the substrate from binding. If this permanently alters the three-dimensional structure of the enzyme, the enzyme is said to **denature**.

Lower temperatures increase the amount of activation energy required (even with the enzyme catalyst) and the rate of reaction is reduced. While lower temperatures slow the rate of reaction, they do not cause the enzyme to denature (Figure 3.2.6).

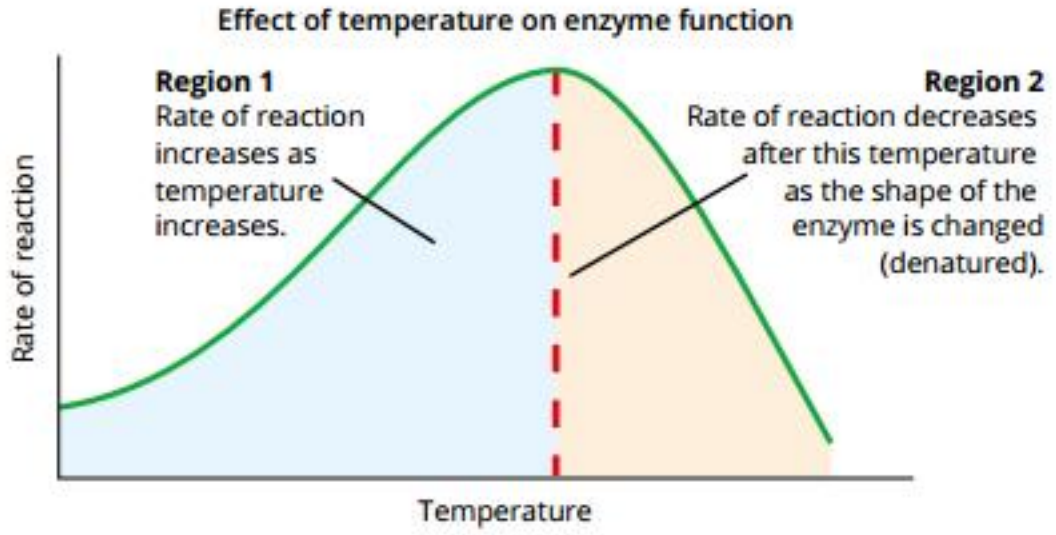

Effect of temperature on enzyme function
Region 1
Rate of reaction increases as temperature increases.
Region 2
Rate of reaction decreases after this temperature as the shape of the enzyme is changed (denatured).
Rate of reaction
Temperature

FIGURE 3.2.6 The rate of reaction increases as temperature increases (Region 1) up to an optimum. The rate of reaction decreases after this temperature because the shape of the enzyme changes or it becomes denatured (Region 2).

pH

Some amino acid side chains can be negatively charged (e.g. glutamic acid) or positively charged (e.g. lysine). This means they are electrostatically attracted to one another, which strengthens the folding of the enzyme's polypeptide chains. However, in extreme pH environments, both excess H^+ (hydrogen) and OH^- (hydroxide) ions can interfere with the positive-to-negative interactions and cause the enzyme to unfold and denature.

Enzymes have very narrow optimal pH environments, as seen in the graphs in Figure 3.2.7. For example, most enzymes in the human body have an optimal pH range of 6–8. However, the pepsin enzyme functions in the stomach to break down proteins in food as a part of the digestion process. Pepsin has very strong hydrogen and ionic bonds between its amino side groups; consequently, it has an optimal pH range around 2.

Inhibitors

Access to the active site on enzymes for substrates can also be impeded by molecules that have a similar active site configuration. This is known as **competitive inhibition** and is shown in Figure 3.2.8. An example of competitive inhibition is the treatment of methanol poisoning with ethanol. Methanol is converted to toxic formaldehyde by the enzyme—alcohol dehydrogenase (ADH). Ethanol competes for the same active site on the ADH enzyme. Thus, ethanol is an antidote sometimes given to a patient with methanol poisoning, such as from homemade distilled fruit spirits. The ethanol will later be oxidised to safe products and finally be eliminated as carbon dioxide and water.

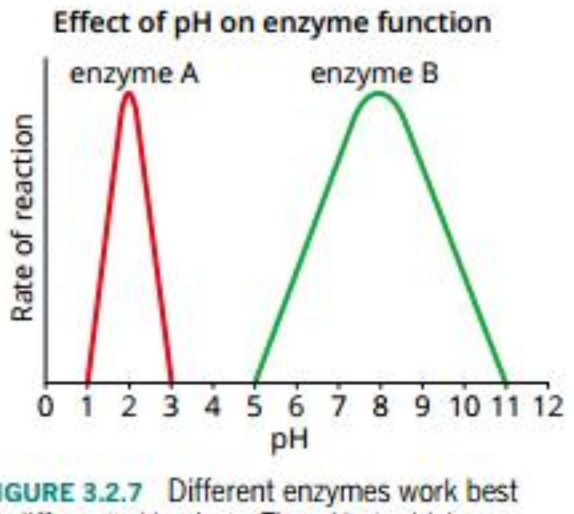


FIGURE 3.2.7 Different enzymes work best at different pH values. The pH at which an enzyme works best is called its optimum pH. For example, intestinal enzymes (like enzyme B) work best under alkaline conditions, thus have an optimum pH of 8. Enzymes that work in the stomach (like enzyme A) have an optimum pH of 2 because the stomach is very acidic.

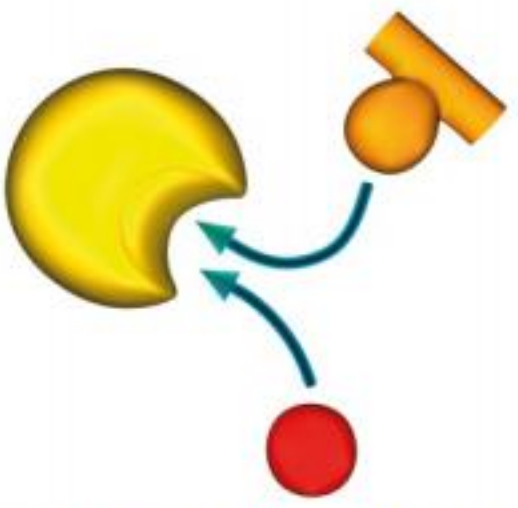


FIGURE 3.2.8 Competitive inhibition is when similarly shaped binding sites of molecules compete for access to an enzyme's active site.

Non-competitive inhibition occurs when a molecule attaches to another site on the enzyme. The interaction of molecular forces causes the enzyme active site to change shape. This effectively blocks the docking of the enzyme's specific substrate to its active site. An example of this is potassium cyanide, which docks on the cytochrome molecules in the mitochondrial membrane. This effectively stops cellular respiration and the major production of ATP.

Concentration

Higher substrate concentration leads to faster reactions controlled by enzymes, until a saturation point is reached with all enzymes available being used (Figure 3.2.9). Increasing the number of enzymes present will only further increase the reaction rate if there is excess substrate present. Past the saturation point, the reaction rate remains stable until either the substrate is used up or the reaction product begins to block substrate access to the enzyme. At this point, all the available active sites are being used.

Control of substrate or enzyme concentration is used by the body to regulate the speed of its biochemical reactions. For example, the products of cellular respiration and photosynthesis reactions are moved away from their production sites in the cell to prevent buildup and inhibition of these reactions. Control of hormone production and passage of nerve impulses by neurotransmitter molecules is regulated this way.

The Data analysis section on page 98 outlines an experimental investigation of enzyme activity and substrate concentration.

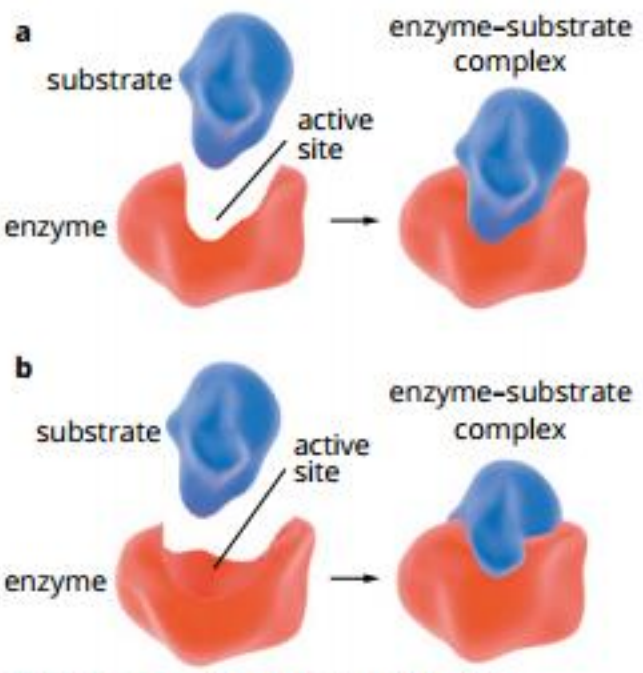


FIGURE 3.2.10 A comparison of the (a) lock-and-key mechanism with the (b) induced-fit model of enzyme action. Notice in the induced-fit model, how the enzyme distorts and fits more tightly around the substrate to form the enzyme–substrate complex.

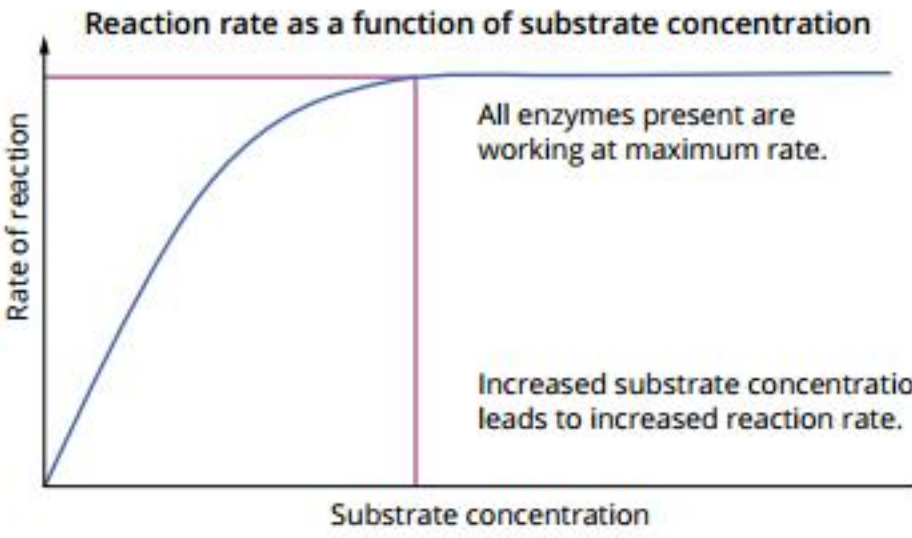

Reaction rate as a function of substrate concentration
Rate of reaction
All enzymes present are working at maximum rate.
Increased substrate concentration leads to increased reaction rate.
Substrate concentration

FIGURE 3.2.9 Increasing the substrate concentration increases the rate of reaction until all the enzymes are working at their maximum capacity.

Lock-and-key versus induced-fit models

For many years, the lock-and-key model was considered the structural mechanism by which enzymes worked (Figure 3.2.10a). Then it was discovered that enzymes are not rigid; when the substrate enters the groove of the active site, the enzyme induces a change in shape to fit itself tightly against the substrate. This brings the amino acid side groups into very close contact with the critical sites on the substrate. After the reaction occurs, the enzyme changes back to its original shape and releases the product. This more accurate model of understanding is known as the induced-fit model and is shown in Figure 3.2.10b.

3.2 Review

SUMMARY

- Enzymes are protein molecules and most are specific to a substrate.
- Enzymes have a groove on their surface containing active sites that interact with the substrate.
- The currently accepted model for enzyme function is the induced-fit model. In this model, the enzyme shapes itself around the substrate thereby reducing the activation energy required for a biochemical process to occur.
- In the earlier lock-and-key model, the enzyme's active site was thought to be a rigid shape which the substrate locked into.
- Enzymes act as catalysts to initiate chemical reactions by lowering the activation energy required for the reaction.
- Enzymes that are deformed in shape, or denatured (e.g. by high temperatures), cannot act as catalysts.
- The rate of reaction is influenced by temperature, pH and the concentrations of substrate and enzyme.
- Some enzymes require cofactors to activate them.
- Most enzymes only effectively operate within a narrow temperature or pH range.
- Inhibitors may also affect the rate at which a substrate can bind to an enzyme.
- Anabolic reactions are ones that require energy input to build up complex molecules. Catabolic reactions release energy when they breakdown complex molecules into simpler ones.

KEY QUESTIONS

Describe

1 Define 'enzyme'.

2 Identify the current model used to explain enzyme function and name the model it replaced.

3 Identify factors that directly affect enzyme function.

4 Distinguish between anabolic and catabolic reactions, giving an example for each.

5 Where in the human body would you expect to find enzymes?

6 Describe how enzymes reduce the activation energy of a reaction.

7 Summarise, in table form, how the four factors of temperature, pH, inhibitors and substrate concentration, affect enzyme reaction rates.

Apply

8 Explain enzyme-substrate specificity and how it relates to enzyme structure.

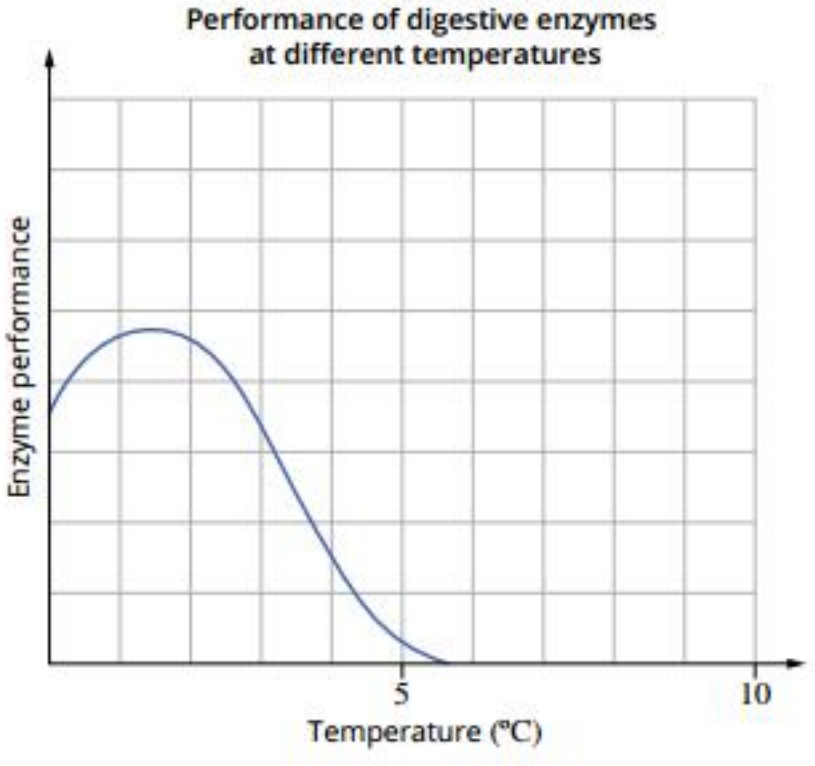
9 Explain why having a protein structure enables enzymes to carry out their function.

10 Explain why the human body maintains a pH around 2 in the stomach and close to 7 in oral saliva.

11 Determine whether enzyme-controlled reactions are catabolic or anabolic when they digest an animal's food intake. Support your answer with an example.

Analyse

12 The following graph shows the effective temperature range of a particular animal's digestive enzyme. Infer what sort of environment this animal could be living in and justify your answer.



continued over page

3.2 Review *continued*

13 Robert O'Hara Burke and William Wills were famous Australian explorers. They were the first Europeans to cross Australia from south to north and nearly back again. It is thought that they perished from a lack of vitamin B_1 (thiamin), an important enzyme cofactor. Near the end of their lives, when they were stranded at Coopers Creek, they subsisted almost entirely on the ground sporocarps (spore cases) of the aquatic fern *Marsilea drummondii,* or nardoo (shown below). They mixed the flour from the sporocarps and nardoo with water to make a type of thin porridge. Nardoo contains the enzyme thiaminase. The local Yandruwandha women also used nardoo in their flour to bake damper. Determine why the thiaminase in the nardoo may have caused the death of Burke and Wills, but not the local Yandruwandha people.

14 Compare the two enzyme models.

3.3 Cellular respiration

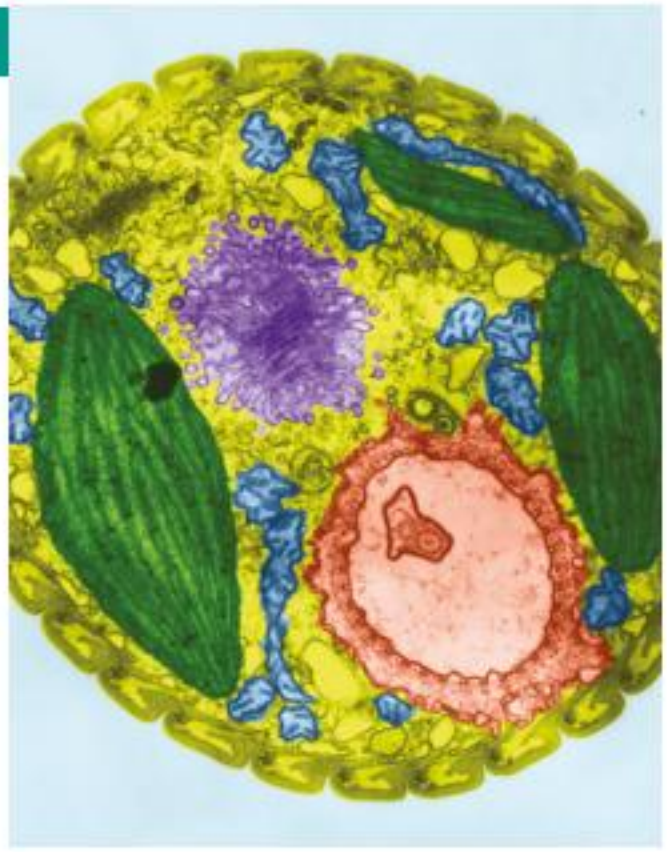
BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- recognise that all life on Earth requires both an energy source and a carbon source
- understand the importance of aerobic cellular respiration reactions in organisms
- understand that cellular respiration is an enzyme-controlled series of chemical reactions and that the reaction sequence known as aerobic respiration (glycolysis, Krebs cycle and electron transfer chain) requires oxygen
- recognise that the overall process can be summarised in a balanced chemical equation:

$$\text{glucose} + \text{oxygen} \rightarrow \text{carbon dioxide} + \text{water} + \text{energy}$$

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + 36\text{–}38ATP$$

- understand that in conditions where there is a lack of oxygen, ATP is produced from glucose by the reaction sequence known as anaerobic respiration (glycolysis with fermentation in plants, yeasts and some bacteria, or lactic acid production in animals)



Cells need energy to do work. The energy used by organisms and their cells is stored in organic compounds. Whether organisms are unicellular or multicellular, or whether they live at the bottom of the ocean or in a rainforest, they all need to take in nutrients and water, exchange gases, obtain energy and remove waste products. Ultimately, most biological systems rely on one source of energy for their survival: sunlight (Figure 3.3.1).

FIGURE 3.3.1 Sunlight is the primary source of energy for biological systems and the organisms within them.

ENERGY VIA ATP

Energy can be defined as the ability to cause change. For example, you are using energy right now to move your eyes to read these words, just as the cells inside your body are currently using energy to transport substances across their membranes. You, like all organisms, are constantly expending energy to make changes.

Energy exists in many forms. The energy in sunlight is solar energy, the heat generated by your body is thermal energy, and when you turn a page, this movement involves kinetic energy. Chemical energy is the potential energy that can be released by a chemical reaction. Chemical energy is stored in the bonds or connections that join atoms together. For example, between atoms of carbon and hydrogen in organic compounds such as glucose, fats and proteins. The cells of all organisms draw upon this chemical energy by breaking compounds down (catabolism). The chemical energy released is then used to make a universal transport energy molecule. This molecule is **adenosine triphosphate**, also known as ATP.

ATP

ATP molecules are composed of three inorganic phosphate groups attached to the nitrogenous base, adenosine. Adenosine is formed when a ribose sugar bonds with the nitrogen-carrying molecule called adenine. The ATP molecule contains two high-energy bonds between the inorganic phosphate groups (Figure 3.3.2). These bonds can be easily broken to release a small 'packet' of energy. These packets of energy are used to carry out all the energy-dependent processes of cells. The number of ATP used at a time depends on how much energy is required.

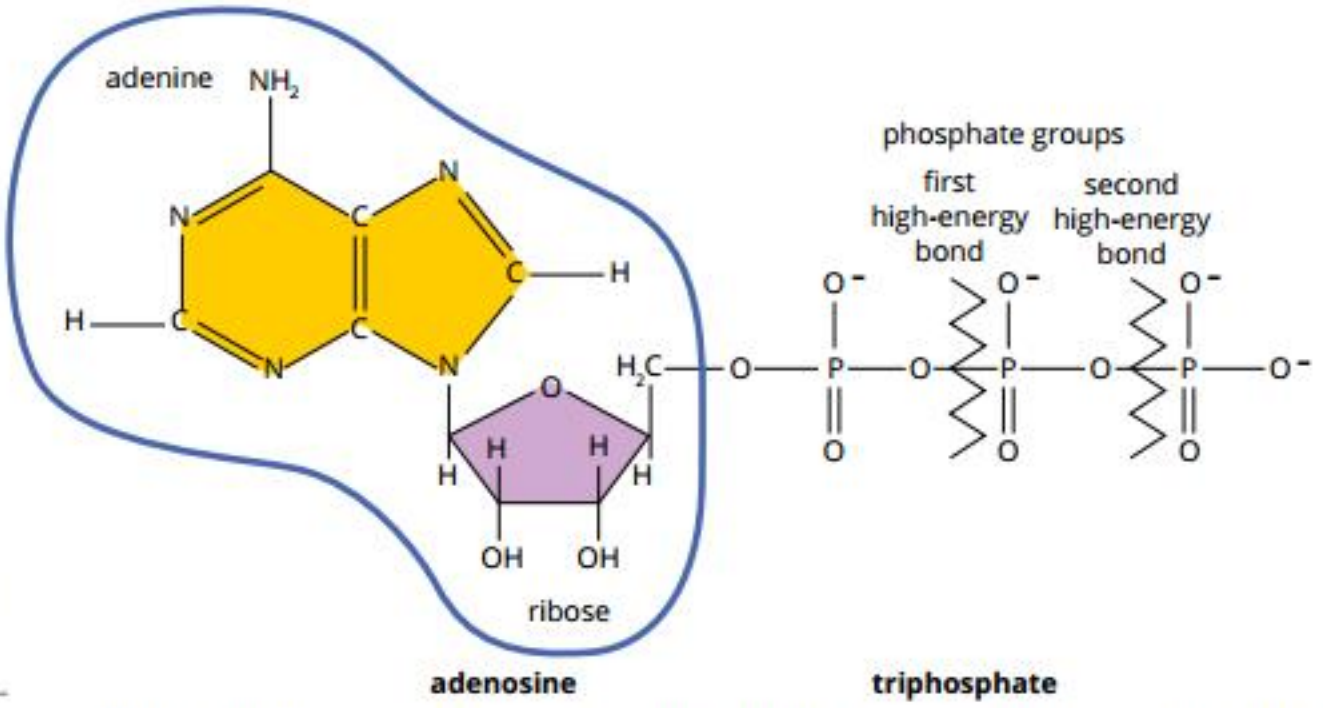


FIGURE 3.3.2 The structure of adenosine triphosphate (ATP). ATP contains the sugar ribose, the nitrogenous base adenine (together forming adenosine) and a chain of three phosphate groups (triphosphate) bonded to it.

When an ATP molecule gives up its energy, it is a catabolic reaction. The ATP splits into a molecule of ADP (adenosine diphosphate) and a molecule of phosphate. This process is reversible because, as an anabolic reaction, the ADP can combine with a phosphate molecule to form an ATP molecule again using energy derived from the breakdown of glucose during cellular respiration (Figure 3.3.3). This recycling process requires much less energy than it would take to make an entirely new ATP molecule. Like all biochemical reactions, the conversions between ATP and ADP are catalysed by their specific enzymes.

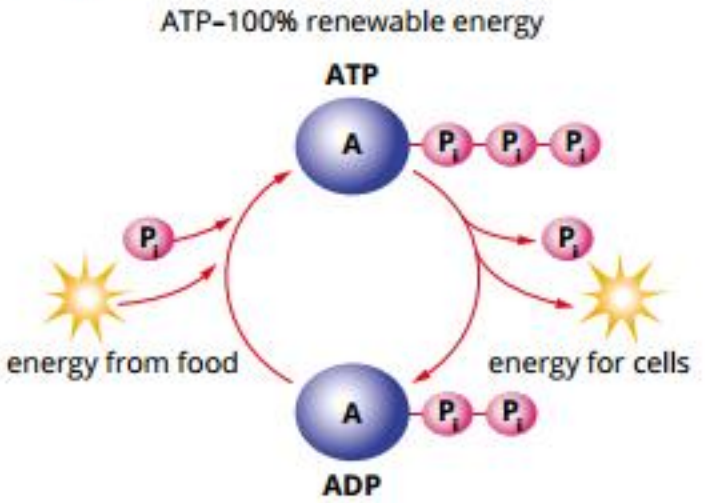


FIGURE 3.3.3 Breaking down ATP releases energy in a catabolic reaction. Cells constantly replace their ATP by adding a spare phosphate onto ADP (adenosine diphosphate) in an anabolic reaction. Enzymes control the synthesis and breakdown of ATP.

CELLULAR RESPIRATION AND PHOTOSYNTHESIS

Organisms can be divided into two groups depending on the strategies they use to obtain organic compounds (Figure 3.3.4). **Autotrophs** obtain organic compounds by converting inorganic matter. They convert carbon dioxide and water to organic glucose molecules in an enzyme-controlled process. Glucose is later converted to other carbohydrates or provides the energy to build other biomolecules. Because they produce all the organic compounds in ecosystems, autotrophs are also called producers. Most autotrophs use photosynthesis. Photosynthesis is covered in more detail in Module 3.4. The typical photosynthetic organisms you might think of are green plants, but there are also cyanobacteria and photosynthetic protists such as algae and *Euglena* (Figure 3.3.5).

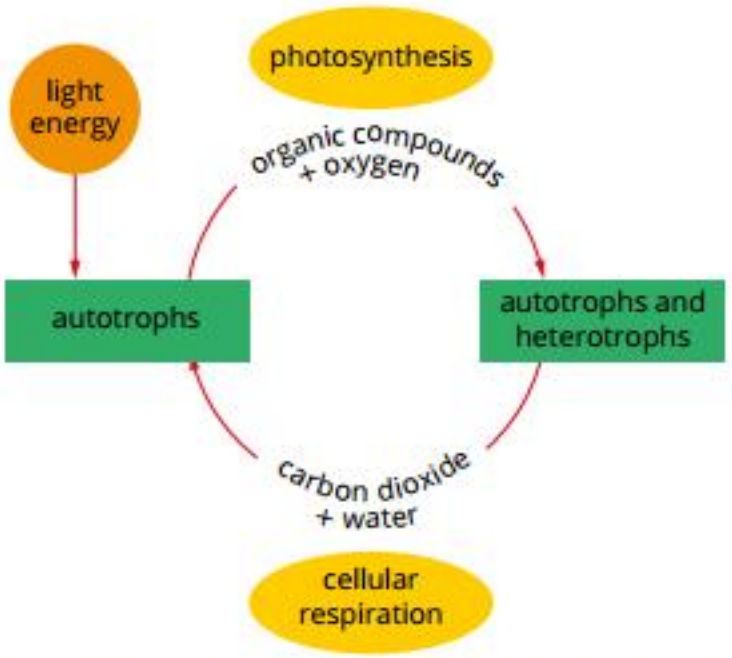


FIGURE 3.3.4 Autotrophic organisms make the organic compounds they require by combining inorganic compounds from their environment. Heterotrophic organisms obtain the organic compounds they require by eating other organisms or products of other organisms.

Heterotrophs ('other feeders') are also called consumers because they are unable to make their own food. Unlike autotrophs, heterotrophs cannot use simple inorganic substances to make organic compounds. Instead, they must obtain the organic compounds they need by consuming other organisms or their products (Figure 3.3.4). All animals and fungi are heterotrophs. Some bacteria and many protozoans are also heterotrophs.

A few organisms are both autotrophic and heterotrophic. In sunlight, the protozoan *Euglena* species, shown in Figure 3.3.6, is photoautotrophic. But when there is no sunlight, they can absorb food from their environment, so they are also heterotrophic.

FIGURE 3.3.6 *Euglena* species are single-celled flagellate protozoans that can obtain energy from both sunlight and other organisms. They are an uncommon example of an organism being both autotrophic and heterotrophic.

FIGURE 3.3.5 Photoautotrophs use light energy to synthesise organic molecules from carbon dioxide and water. (a) On land, plants are the main photoautotrophs. In aquatic environments, the main photoautotrophs are (b) algae such as this kelp and (c) prokaryotes called cyanobacteria.

CELLULAR RESPIRATION

All cells in both autotrophs and heterotrophs need energy to function. They obtain this by releasing energy from organic compounds through a series of biochemical pathways; **glycolysis** is the first of these. Cellular respiration is the name given to the combination of biochemical pathways that together release energy from glucose, the structure of which is shown in Figure 3.3.7.

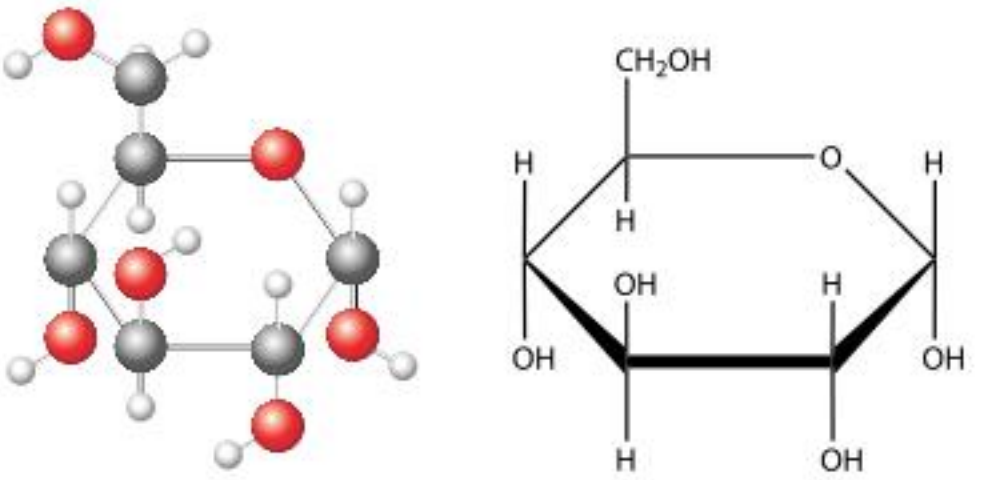


FIGURE 3.3.7 Glucose is the primary energy source for cellular respiration. In the ball-and-stick model, the carbon atoms are black. In the molecular diagram of glucose, there is a carbon atom at each angle in the ring, unless labelled otherwise. This is standard practice for drawing organic molecules built from carbon.

FIGURE 3.3.8 Complex carbohydrates are broken down by the digestive system into simple sugars, such as glucose.

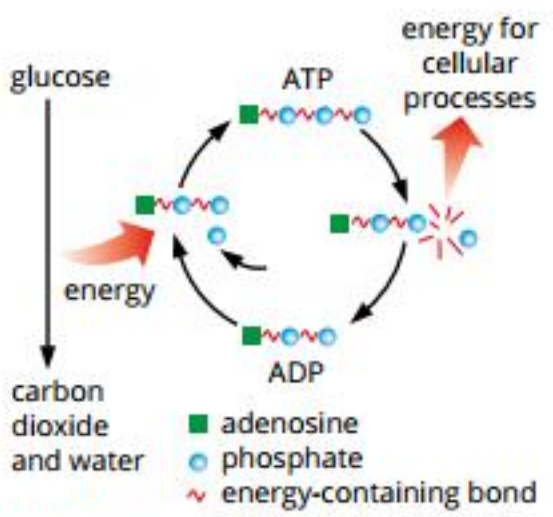


FIGURE 3.3.9 Energy from glucose is transferred in the synthesis of ATP from ADP and phosphate. The energy is stored in the phosphate bond. When the bond is broken, the energy is released to drive cellular processes. The ADP and phosphate are recycled.

ENERGY FROM GLUCOSE

Most cells use glucose as their immediate source of energy. Glucose can be obtained by breaking down complex carbohydrate foods such as those shown in Figure 3.3.8. It is possible to extract energy from other organic molecules such as lipids and proteins, but it is a more complicated process with less energy reward. Some molecules like lipids must be converted into glucose first; others are broken down to small molecules that can also be used in the cellular respiration pathways.

Each reaction in the cellular respiration pathway is catalysed by a particular enzyme.

As described above, the energy released from glucose through cellular respiration is used to generate molecules of adenosine triphosphate (ATP) (Figure 3.3.2). The energy is transferred when ATP is formed from ADP and inorganic phosphate (P_i), and it is stored in the bond between ADP and phosphate. This can be represented as:

$$ADP + P \rightarrow ADP\sim P_i \text{ (ATP)}$$

The '~' represents the high-energy bond in which the energy is stored.

When the high-energy bond in ATP is broken, energy is released for use in the many energy-demanding processes that occur in the cell. The transfer of energy is summarised in Figures 3.3.3 and 3.3.9.

Aerobic cellular respiration requires oxygen and consists of the three interconnected biochemical pathways known as:

- **glycolysis**
- the **Krebs cycle** (also called the citric acid cycle)
- the **electron transfer chain** (also called the electron transport chain).

Anaerobic cellular respiration also involves glycolysis. In plant and yeast cells, the product of glycolysis undergoes a **fermentation** pathway in the absence of oxygen.

GLYCOLYSIS

Glycolysis involves the breakdown of sugar (glucose) to release energy. The word 'glycolysis' comes from the Greek words '*glykys*', which means 'sweet', and '*lysis*', which means 'to loosen or break down'.

In the process of glycolysis, a small amount of energy is released. Glycolysis is the first stage of cellular respiration and occurs in the cytosol of the cell. It results in the net production of two ATP molecules and two **NADH (nicotinamide adenine dinucleotide)**. Both types of molecules, ATP and NADH, act as energy carriers. During glycolysis, the glucose (a six-carbon molecule) is broken down to two three-carbon molecules called **pyruvate** (or pyruvic acid) in a catabolic reaction.

Aerobic and anaerobic respiration pathways

After glycolysis, cellular respiration can be diverted into one of two biochemical pathways depending on the availability of oxygen. These are **aerobic respiration** and **anaerobic respiration**. Here you will learn some of the details of the biochemical reactions involved, where these processes occur in the cell, and the different amounts of energy released in each pathway.

AEROBIC RESPIRATION

If oxygen is available, most eukaryotic organisms use it to release energy from glucose in the process of aerobic respiration. As noted above, aerobic respiration comprises three stages: glycolysis, the Krebs cycle and the electron transfer chain. Each stage occurs in a different location of the cell as shown in Figures 3.3.10 and 3.3.11. The latter two stages occur in the mitochondria and require oxygen. Glycolysis occurs in the cytosol and breaks down glucose molecules regardless of oxygen being present or not. Note that, although the overall pathway of aerobic cellular respiration produces up to 38 molecules of ATP, only two ATP molecules are produced by glycolysis.

The overall balanced chemical equation for aerobic cellular respiration is shown below:

glucose + oxygen → carbon dioxide + water + energy

$$C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + 36\text{–}38ATP$$

Mitochondria

Mitochondria are often referred to as the powerhouses of the cell. They are the sites of the Krebs cycle and the electron transfer chain—two of the three processes that comprise aerobic respiration. If the cell has a supply of oxygen, the pyruvate formed from glycolysis in the cytosol passes into the mitochondria, where it is further broken down through a series of biochemical steps into carbon dioxide and water. As always for biochemical reactions, these steps are catalysed by enzymes.

Mitochondria are slightly smaller than chloroplasts. Inside the outer membrane, there is a folded inner membrane structure. The folded structures of the inner membrane are called cristae. Between the cristae is the mitochondrial matrix, as shown in Figures 3.3.10 and 3.3.12.

The more intricately folded a membrane, the greater the surface area it has available for enzyme attachment. This is of great importance as you will see in the next two stages of aerobic respiration—the Krebs cycle and the electron transfer chain—both of which occur in the mitochondria.

Krebs cycle

The second important stage of aerobic respiration occurs after glycolysis and is called the Krebs cycle. It takes place in the mitochondrial matrix. The Krebs cycle is a series of eight reactions, each catalysed by a different enzyme. The pyruvate formed from glycolysis diffuses from the cytosol through the outer membrane of the mitochondria and is then moved by active transport through the inner membrane. Moving pyruvate actively into the mitochondria uses ATP generated by glycolysis.

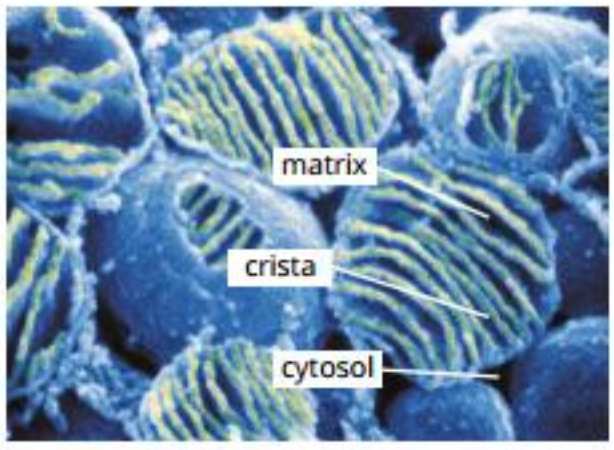


FIGURE 3.3.10 The first stage of aerobic cellular respiration (glycolysis) occurs in the cytosol. The second stage (the Krebs cycle) occurs in the matrix of the mitochondria. The last stage (the electron transfer chain) occurs on the cristae of the mitochondria.

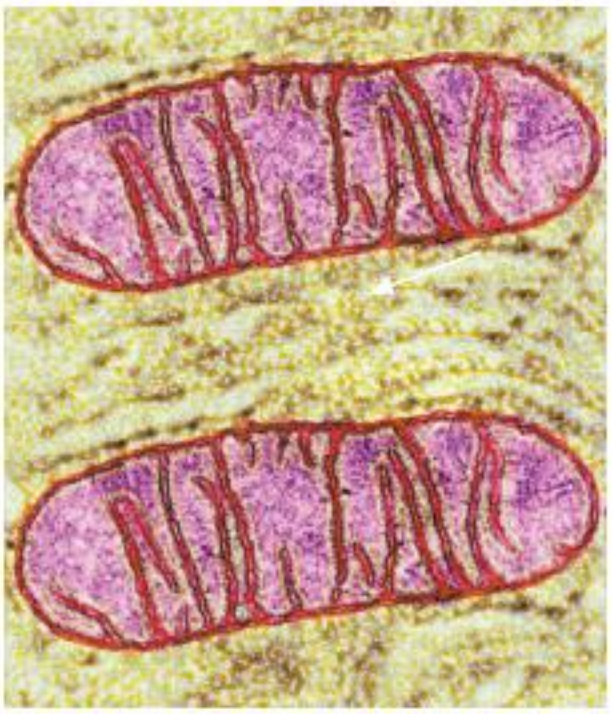


FIGURE 3.3.12 This coloured transmission electron micrograph of a mitochondrion shows the inner membrane (cristae) in red and the matrix fluid in purple.

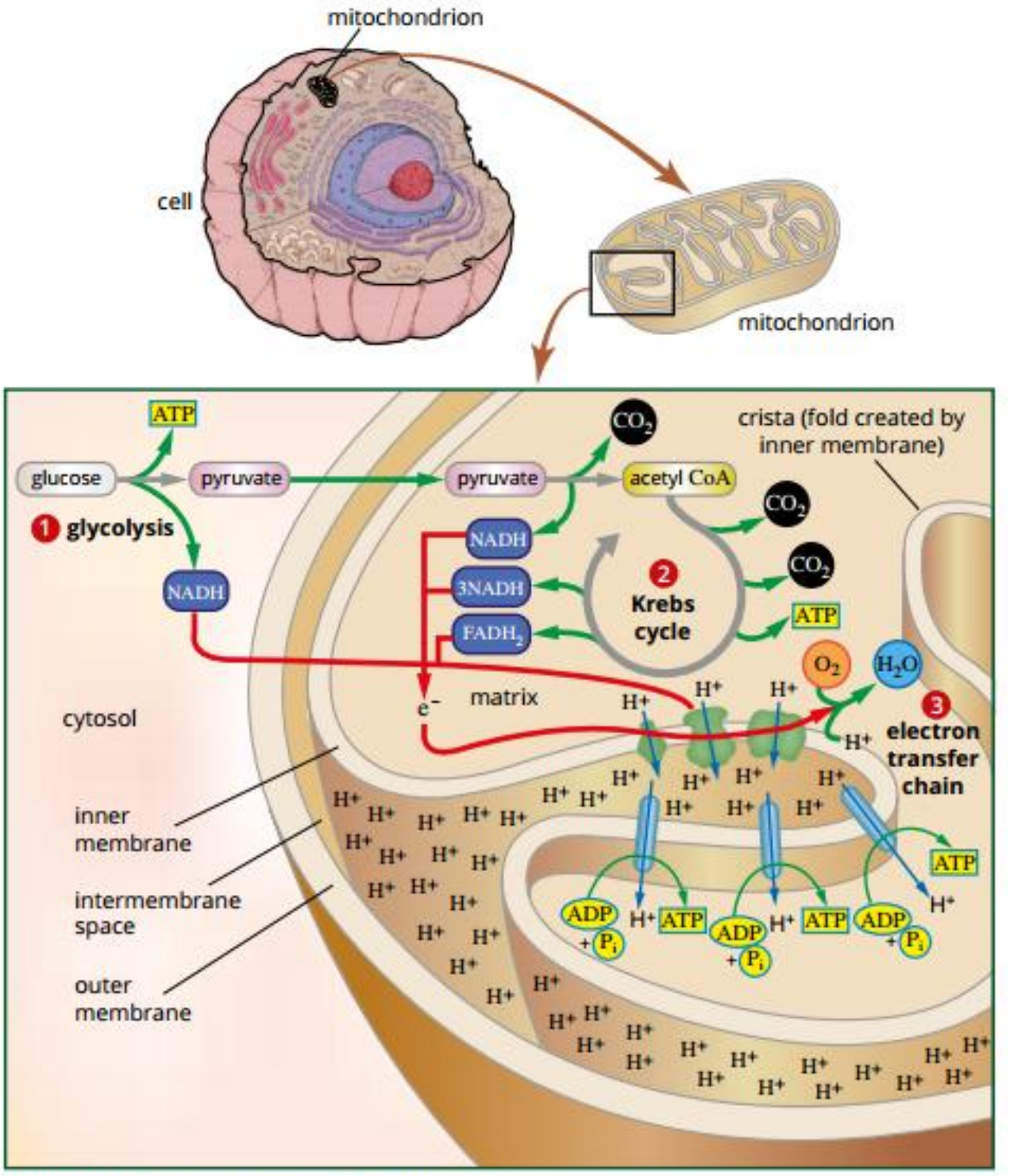


FIGURE 3.3.11 A summary of aerobic cellular respiration

When the pyruvate, a three-carbon molecule, is in the mitochondrial matrix, it is converted into acetyl coenzyme A (acetyl CoA), a two-carbon molecule, which is the substrate for the first of a series of reactions that make up the Krebs cycle. In the formation of acetyl CoA from pyruvate, one carbon dioxide molecule is formed. In addition, in one turn of the Krebs cycle two carbon dioxide molecules are formed. That is a total of three molecules of carbon dioxide formed for every pyruvate molecule and six molecules of carbon dioxide for every glucose molecule metabolised.

During the reactions of the Krebs cycle, energy is transferred to energy-carrying coenzymes such as Nicotinamide Adenine Dinucleotide (NAD), FAD and ADP during the formation of NADH, Flavin Adenine Dinucleotide (**$FADH_2$**) and ATP. This is shown in Figures 3.3.11 and 3.3.13.

Electron transfer chain

The overall purpose of the electron transfer chain (also known as the electron transport chain) is to move sub-atomic protons and electrons across a membrane as a system for generating ATP. For this process, oxygen is essential because it picks up electrons at the end of the chain. If oxygen is not available, the electron transfer chain stops.

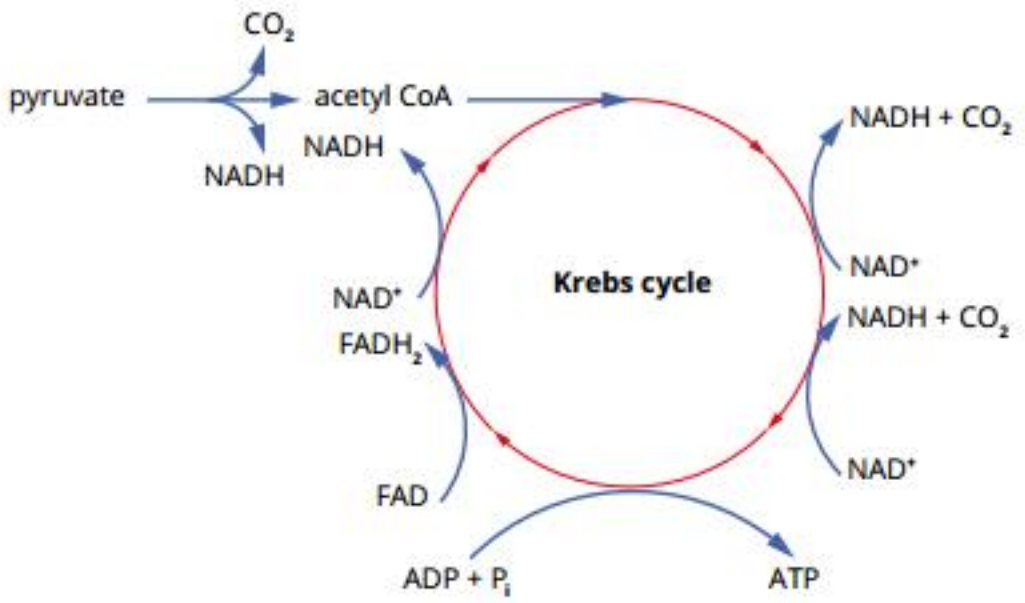


FIGURE 3.3.13 The three-carbon molecule pyruvate produced in glycolysis is converted to a two-carbon molecule acetyl CoA with the production of a carbon dioxide molecule. The acetyl CoA then enters the Krebs cycle where it results in ATP, $FADH_2$, and more CO_2 and NADH being formed.

Embedded in the cristae (folds) of the inner membrane of the mitochondria are a number of protein complexes, including enzymes and cytochromes, as shown in Figure 3.3.14. These complexes form an interconnected series of reactions that together make up the electron transfer chain, which is the third stage of aerobic cellular respiration.

Energy-carrying molecules from the Krebs cycle feed into the electron transfer chain. NADH is converted back to NAD^+ by interacting with the first complex at the beginning of the electron transfer chain, and $FADH_2$ is converted back to FAD by interacting with the second complex.

The hydrogen ions (H^+) originating from the conversion of NADH and $FADH_2$ are moved into the intermembrane matrix of the mitochondria and the electrons are transferred along the chain. The energy obtained in this process is used to make ATP from ADP. At the end of the electron transfer chain, hydrogen ions and electrons combine with oxygen to form water.

The electron transfer chain forms 26–34 molecules of ATP, using the energy that was contained originally in each glucose molecule.

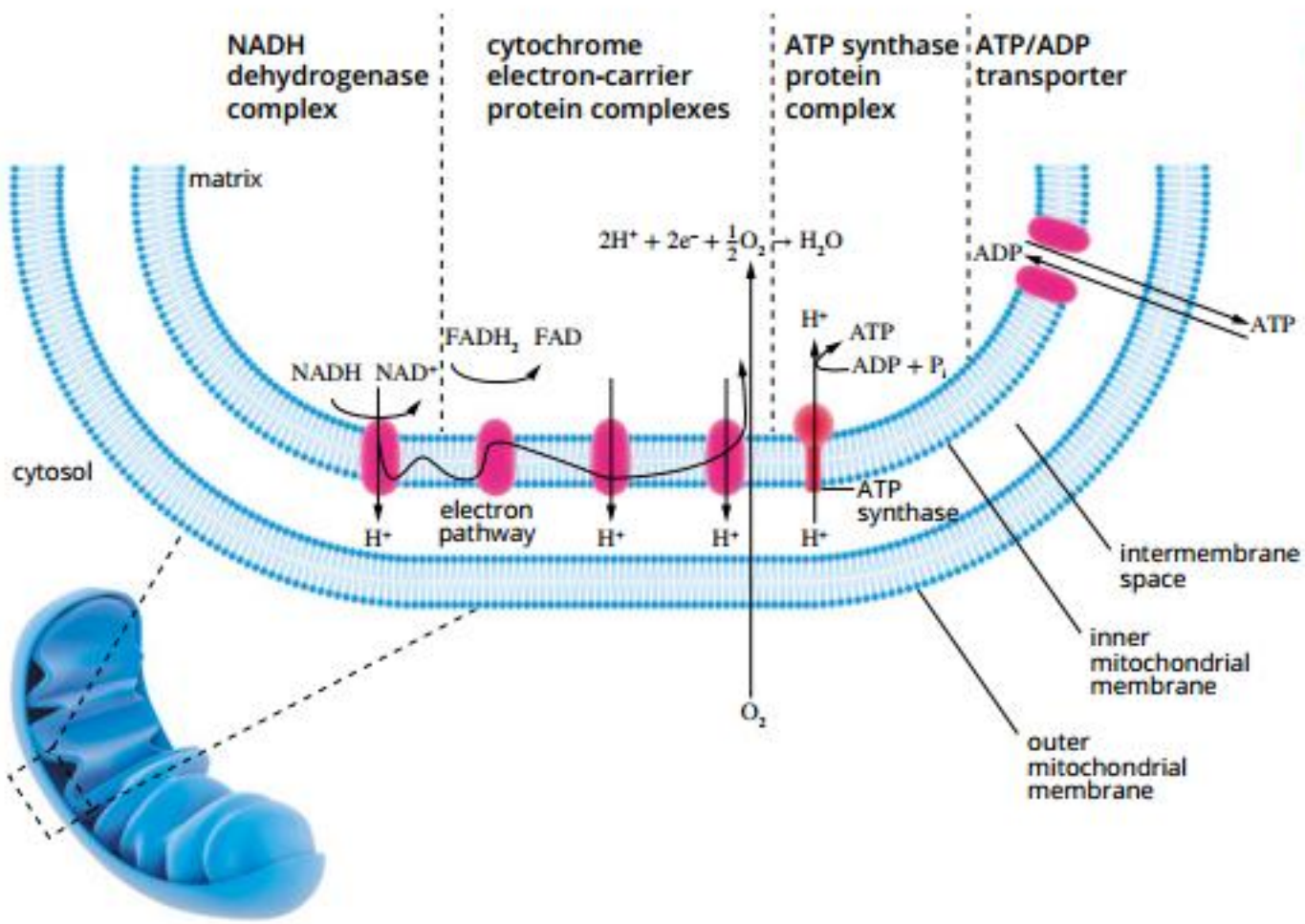


FIGURE 3.3.14 The inner mitochondrial membrane has several embedded proteins that pass along the energy and H^+ ions from carriers (NADH and $FADH_2$) during the electron transfer chain. The final enzyme in the pathway, ATP synthase, generates large amounts of ATP.

ATP yield in aerobic respiration

If you add up the ATP molecules formed at each stage of aerobic respiration, you find that for every molecule of glucose metabolised, 30–38 ATP molecules can be formed:

- 2 ATP from glycolysis
- 2 ATP from the Krebs cycle
- 26–34 ATP from the electron transfer chain.

It had been generally accepted that 36–38 ATP molecules are produced overall from the breakdown of one glucose molecule by aerobic respiration. However, some research now indicates there may be more variation than previously thought, with some cells and tissues being more efficient than others. A revised range of 30–38 ATP may be more appropriate.

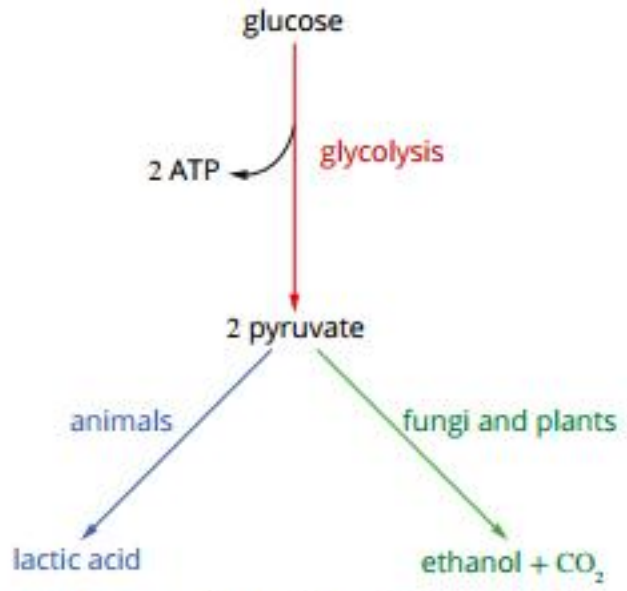


FIGURE 3.3.15 Anaerobic respiration occurs in the cytosol of cells when oxygen is not available.

ANAEROBIC RESPIRATION

If there is little or no oxygen available, a eukaryotic organism can still release some energy from glucose through the anaerobic pathway known as **fermentation**. In animals, **lactic acid** fermentation is the name of the process that most often occurs in the cytoplasm of cells after glycolysis when oxygen is in short supply. In yeast and plants, a different type of fermentation occurs—ethanol fermentation. Figure 3.3.15 shows the products of these two types of anaerobic respiration.

COMPARING AEROBIC AND ANAEROBIC RESPIRATION

Anaerobic respiration pathways in eukaryotes produce only two ATP molecules per glucose molecule during glycolysis, whereas aerobic respiration produces 30–38 ATP molecules per glucose molecule during glycolysis, the Krebs cycle and the electron transfer chain. Therefore, aerobic respiration is much more efficient in supplying the cell with energy. The organic products of anaerobic respiration (lactic acid from animals and alcohol from yeast) still contain a lot of energy and both can be further metabolised to release more energy. Figure 3.3.16 and Table 3.3.1 compare the processes of aerobic and anaerobic respiration.

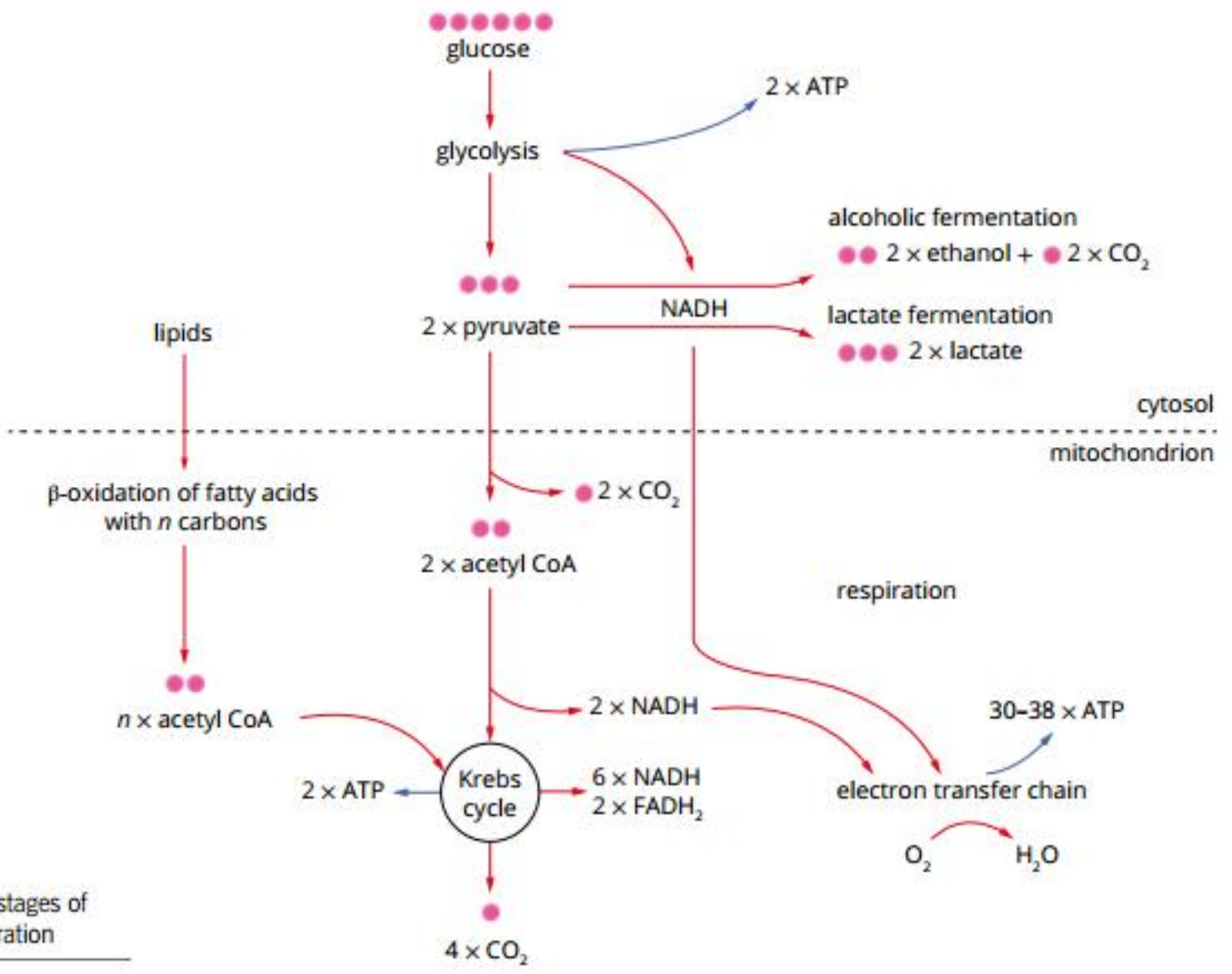


FIGURE 3.3.16 An overview of the stages of aerobic and anaerobic cellular respiration

TABLE 3.3.1 Summary of cellular respiration

	Aerobic	Anaerobic
location	cytosol and mitochondria	cytosol
reactants	glucose and oxygen	glucose
products	carbon dioxide and water	• lactic acid (animals) • ethanol and CO_2 (plants/fungi)
energy output (per glucose molecule)	30–38 ATP total	2 ATP total

FACTORS AFFECTING THE RATE OF CELLULAR RESPIRATION

Cellular respiration is affected by a number of factors including temperature, glucose availability and oxygen concentration. Like any other enzyme-controlled reaction, the availability of reactants is a factor and an optimum temperature for the fastest rate of aerobic respiration (Figure 3.3.17). For humans, 36–38°C gives the fastest rate of cellular respiration.

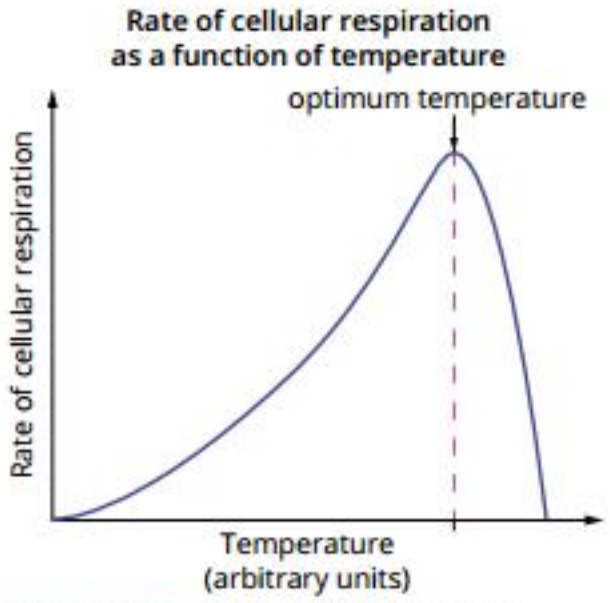

Rate of cellular respiration as a function of temperature
optimum temperature
Rate of cellular respiration
Temperature (arbitrary units)

FIGURE 3.3.17 The relationship between temperature and the rate of cellular respiration. As the temperature increases towards the optimum range, the rate of cellular respiration speeds up. At the optimum temperature, cellular respiration occurs at the maximum rate. At temperatures above the optimum temperature, the rate of cellular respiration rapidly decreases due to the impact of high temperatures on enzyme performance.

SUMMARY OF CELLULAR ENERGY TRANSFORMATIONS

Table 3.3.2 Summarises cellular energy transformations. Photosynthesis is listed here for comparison, and it will be studied in more detail in the next Module 3.4.

TABLE 3.3.2 Summary of cellular energy transformations

	Photosynthesis	Cellular respiration	
		Aerobic	Anaerobic
location	chloroplasts	cytosol and mitochondria	cytosol
reactants	water and carbon dioxide	glucose and oxygen	glucose
products	glucose, oxygen and water	water and carbon dioxide	• lactic acid (animals) • ethanol and carbon dioxide (plants/fungi)
carriers	$NADP^+$	NAD^+ and FAD	NAD^+
ATP produced	18	30–38	2

3.3 Review

SUMMARY

- Autotrophs use energy and inorganic molecules from the physical environment to produce the organic compounds they need.
- Most autotrophs, including plants and algae, are photosynthetic. That is, they use solar energy for producing energy-rich organic compounds.
- Heterotrophs, including animals, fungi and some bacteria and protists, obtain organic compounds by eating other organisms or their products.
- ATP is the main chemical energy transport molecule used by all living organisms.
- Both autotrophs and heterotrophs use the cellular respiration reaction to produce ATP molecules for energy supply.
- Glucose is the primary energy source for cellular respiration.
- The energy released in the breakdown of glucose is carried by ATP, $FADH_2$ and NADH.
- Aerobic cellular respiration has three stages: glycolysis, the Krebs (citric acid) cycle and the electron transfer chain (also called the electron transport chain).
- Two ATP molecules (per glucose molecule) are produced during glycolysis (the first stage of cellular respiration).
- Mitochondria contain folded inner membranes (cristae) and fluid (matrix).
- Glycolysis occurs in the cytosol and yields two ATP. Glucose is converted into two pyruvate molecules.
- The Krebs cycle occurs in the matrix of the mitochondria and yields another two ATP. Pyruvate is broken down into carbon dioxide and acetyl CoA.
- The electron transfer chain occurs in the cristae of the mitochondria and yields 26–34 ATP. Oxygen accepts the hydrogen ions that are used to generate a large amount of energy.
- In animals, the product of anaerobic respiration (without oxygen) is lactic acid. In yeast, the products of anaerobic respiration are ethanol and carbon dioxide.
- Aerobic respiration is much more efficient than anaerobic respiration. Aerobic respiration produces 30–38 ATP per glucose molecule; anaerobic respiration only yields two ATP per glucose molecule.
- Aerobic cellular respiration is affected by temperature, glucose concentration and oxygen concentration.
- When the temperature is above or below the optimum range, the rate of cellular respiration is slower.
- Glucose is a substrate of glycolysis; therefore, an increase in glucose availability increases the rate of cellular respiration.
- Oxygen is a substrate of the electron transfer (transport) chain; therefore, an increase in oxygen concentration increases the rate of aerobic cellular respiration.

KEY QUESTIONS

Describe

1 Explain what is meant by 'chemical energy'.

2 List three types of organic compounds that can be used as a source of energy for cellular respiration.

3 Recall and write the word equation and the balanced chemical equation for the overall reaction of cellular aerobic respiration.

4 Describe what happens when an inorganic phosphate group (P_i) is broken off from a molecule of ATP.

5 Recognise if the glycolysis stage of cellular respiration occurs regardless of the presence or absence of oxygen.

6 Complete the following table about the different energy releasing processes within cells.

Process	Location in cell	Products formed (per glucose molecule)	No. of ATP molecules produced (per glucose molecule)
glycolysis			
Krebs cycle			
electron transfer chain			
fermentation (anaerobic respiration)			

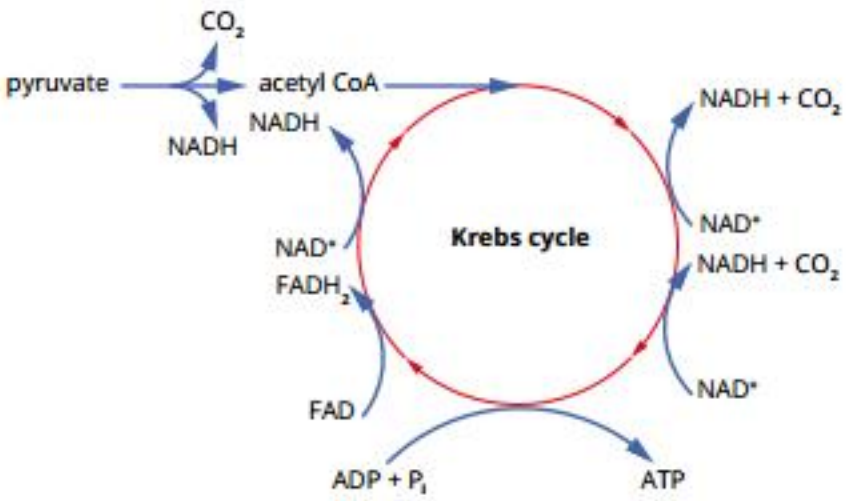
7 Recall the purpose for cells to form ATP molecules.

8 Mitochondria are the site of aerobic respiration in a cell. Draw a labelled diagram of one mitochondrion clearly showing where each of the three stages takes place.

Apply

9 Explain the advantage to a cell of converting pyruvate to lactic acid or ethanol when this yields no extra ATP.

10 The following diagram shows the chemical changes that occur during the Krebs cycle for one molecule of pyruvate.



Use the information in the diagram and your understanding of cellular respiration to answer the following questions:

- **a** Recall how many molecules of ATP are formed in the Krebs cycle from one molecule of glucose.
- **b** Recall how many H^+ ions are loaded onto carriers during one turn of the Krebs cycle.
- **c** **i** State how many molecules of CO_2 are produced during one turn of the Krebs cycle.
  - **ii** Describe the stage that produces the remainder of the CO_2.
- **d** Explain why the electron transfer chain cannot occur without the Krebs cycle.

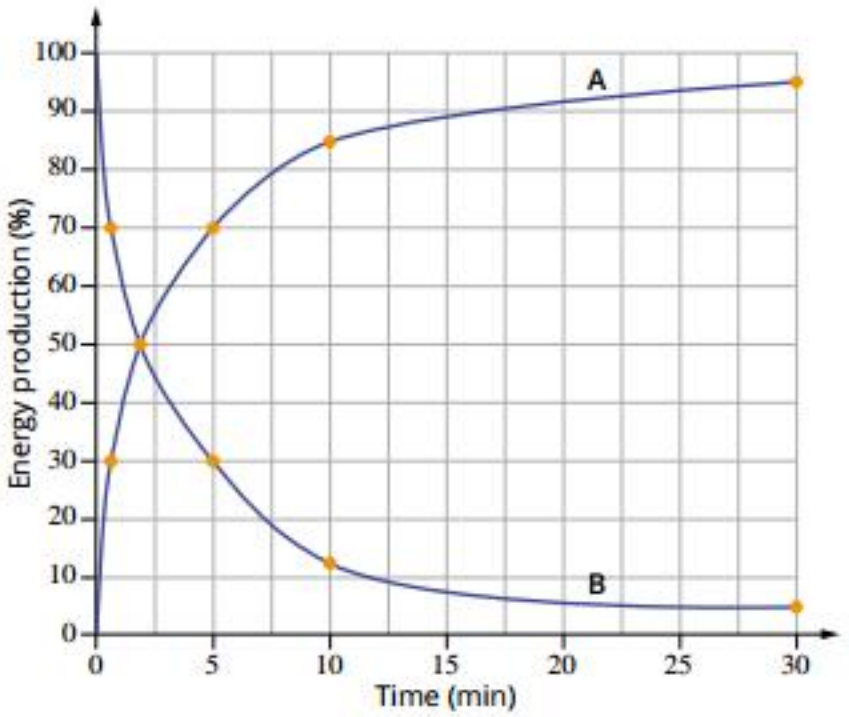
11 Why is ATP production given as a range of numbers?

12 Discuss the value of biomolecules like enzymes, ATP and NADH being reused, whereas other molecules are synthesised for a single use only.

Analyse

13 Compare lactic acid and ethanol fermentation.

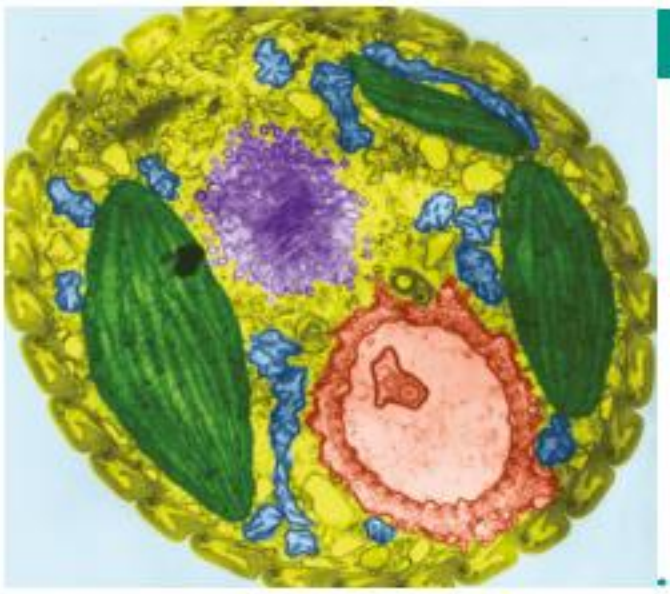
14 The graph shows the contributions of the two energy-producing pathways to physical activity.



Athletes competing in sports requiring short-term power output, such as sprinting, obtain most of their ATP from the anaerobic pathway, but athletes requiring sustained energy use aerobic respiration to meet most of their energy needs.

- **a** Interpret which graph (A or B) is most likely to represent ATP production by a sprinter.
- **b** Determine why anaerobic respiration cannot supply the energy needs of athletes in events requiring energy over a sustained period of time.

3.4 Photosynthesis



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that the process of photosynthesis is an enzyme-controlled series of chemical reactions that occurs in the chloroplast in plant cells and uses light energy to synthesise glucose
- describe the process of photosynthesis in terms of the light-dependent reactions and light-independent reactions
- recognise that the overall process can be summarised in a balanced chemical equation:

$$\text{carbon dioxide + water} \xrightarrow{\text{light energy}} \text{glucose + oxygen + water}$$

$$6CO_2 + 12H_2O \xrightarrow{\text{light energy}} C_6H_{12}O_6 + 6O_2 + 6H_2O$$

FIGURE 3.4.1 Plants trap the energy of sunlight and convert it using photosynthesis into chemical energy.

Simple experiments show that when plants have light, water and carbon dioxide, they make glucose in their green parts, such as leaves. They trap the energy of sunlight and convert it into chemical energy, which they store in the bonds of glucose molecules. This enzyme-controlled process is photosynthesis (*photo*, meaning 'light', and *synthesis*, meaning 'putting together'). All photosynthetic organisms, from single-celled algae to the largest trees, produce glucose in the same way (Figure 3.4.1).

PHOTOSYNTHESIS

Photosynthesis is sometimes called 'carbon fixation' because carbon atoms from the air are incorporated ('fixed') into organic molecules. Photosynthesis involves two stages: a light-dependent stage and a light-independent stage. Each stage involves a series of biochemical reactions, often referred to as a biochemical pathway (or metabolic pathway). Each reaction in the pathway is catalysed (accelerated) by a particular enzyme. Figure 3.4.2 illustrates the process of photosynthesis.

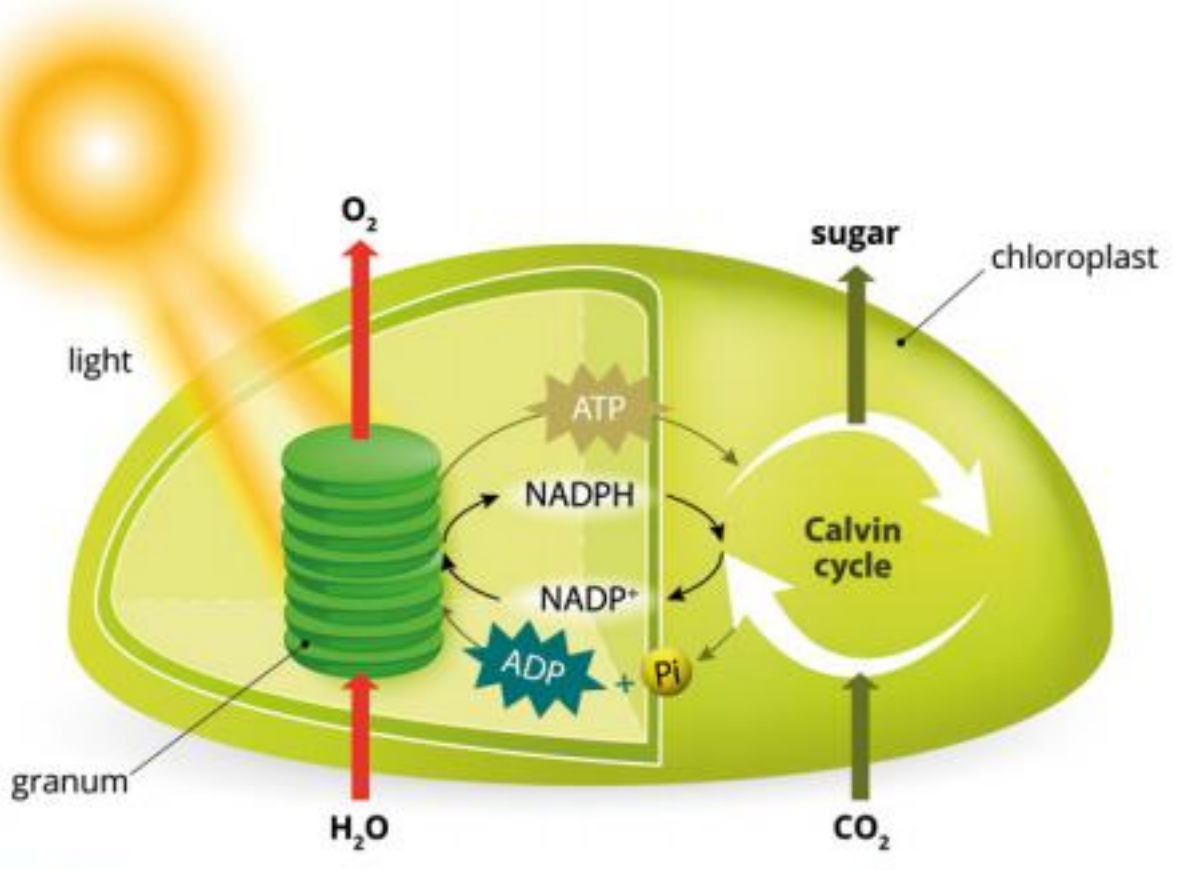


FIGURE 3.4.2 An overview of photosynthesis showing the processes and the products

The glucose formed in photosynthesis may be:

- used as an immediate source of energy by the plant
- stored by the plant as starch for later conversion back to glucose and then used as a source of energy
- used as a chemical starting point for the synthesis of complex compounds, such as cellulose and proteins.

The oxygen formed in photosynthesis may be used for aerobic cellular respiration by the plant or released into the atmosphere.

The reactions that occur during photosynthesis can be summarised by these overall equations:

Word equation: carbon dioxide + water $\xrightarrow{\text{light energy}}$ glucose + oxygen + water

Chemical equation: $6CO_2 + 12H_2O \xrightarrow{\text{light energy}} C_6H_{12}O_6 + 6O_2 + 6H_2O$

CHLOROPLAST STRUCTURE

As you will recall from Chapter 2, the cells of eukaryotic autotrophs, such as plant cells, have specialised organelles called chloroplasts. These lens-shaped organelles contain an outer and an inner membrane, which together regulate the movement of materials into and out of the organelle. Inside these membranes is a fluid matrix called stroma and a highly complex inner thylakoid membrane system increasing the surface area for enzyme-controlled reactions. The thylakoid membranes fold to form flat hollow discs, which form stacks called grana (Figure 3.4.3). Each granum looks like a stack of coins. Linking the grana are flat membrane sheets called lamellae.

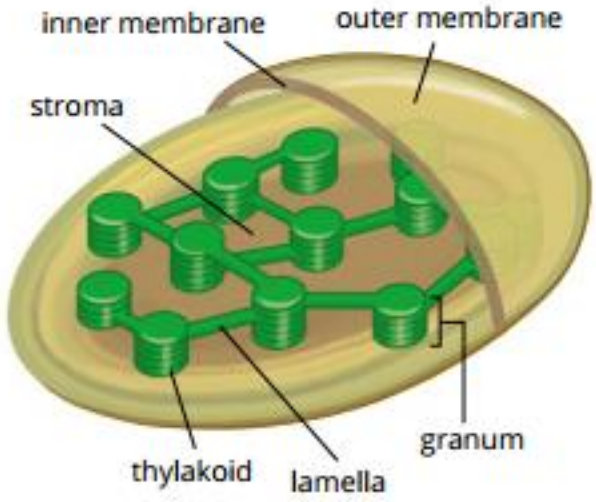


FIGURE 3.4.3 A three-dimensional model of the structure of a chloroplast

Photosynthetic pigments

Photosynthetic pigments are coloured substances that collect the light energy that is used in photosynthesis. The green pigment in chloroplasts is chlorophyll. It is the most abundant and visible photosynthetic pigment found in plants.

Figure 3.4.4a shows you that the rate of photosynthesis is highest under red light (650–700 nm) and lowest under green (500–600 nm). We can explain this using Figure 3.4.4b, because chlorophyll absorbs many of the colours that make up the spectrum of white light but it reflects green light. Plants appear green to us because they reflect more green light than they absorb, whereas other colours are mostly absorbed. The spectrum (Figure 3.4.4b) shows clearly that red, blue and violet light are all strongly absorbed, while yellow is absorbed to a lesser extent and green is not absorbed at all by a green leaf. Note that there are some different types of chlorophyll with slightly different molecular shapes and different light-absorption spectra. The spectra for chlorophyll types a, b and c are shown in Figure 3.4.4b.

The different types of chlorophyll, and other pigments called carotenoids, are all involved in capturing different wavelengths from the white light spectrum. When chlorophyll is broken down in some plants in autumn, the colours of the carotenoids (red, orange and yellow) are no longer masked by the green chlorophyll and are exposed. This gives autumn leaves the colours shown in Figure 3.4.5.

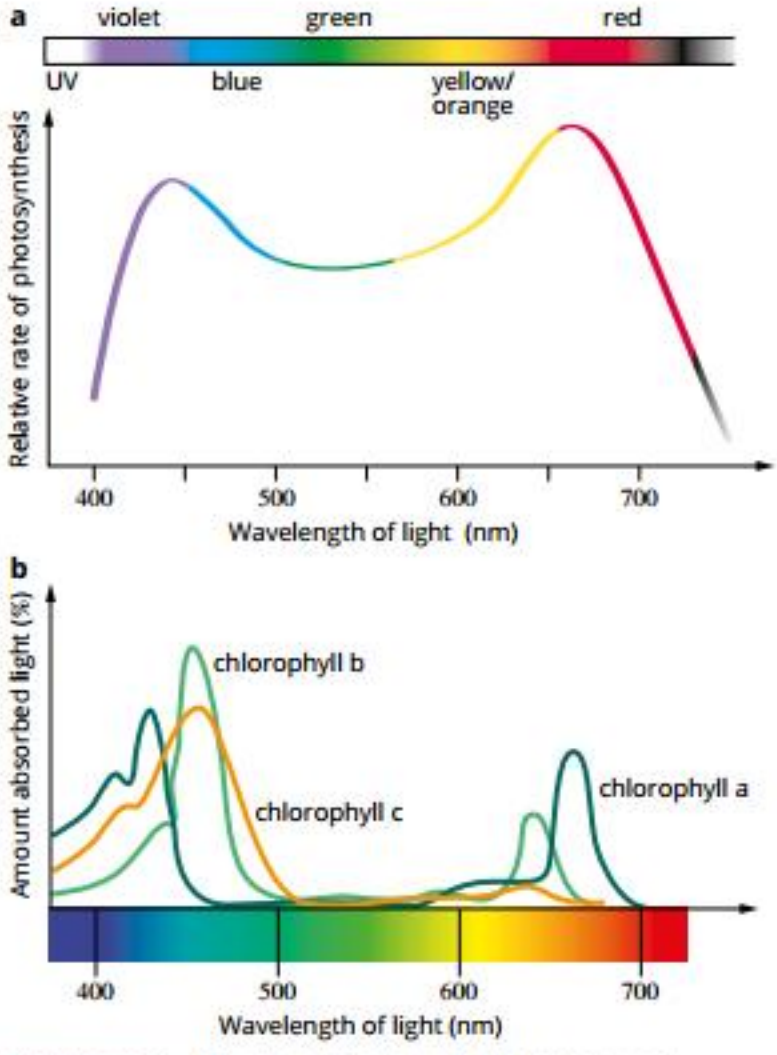


FIGURE 3.4.4 The rate of photosynthesis (a) occurs at different rates in light of different wavelengths. The pattern of photosynthetic activity is very similar to (b) the absorption spectrum of chlorophyll.

FIGURE 3.4.5 Deciduous plants such as the red maple decrease chlorophyll production in autumn, before losing their leaves. The red, yellow, orange or brown colours of carotenoids are visible when there is no green-coloured chlorophyll in the leaves.

LIGHT-DEPENDENT REACTIONS

The first stage of photosynthesis involves biochemical pathways known as the light-dependent reactions. These reactions occur on the thylakoid stacks (grana) inside the chloroplast, where chlorophyll and the enzymes involved are located. The reactions can only take place in the presence of light.

First, light energy is absorbed by chlorophyll. The energy is then used to split water (H_2O) to produce oxygen (O_2), and to form the energy-carrying molecules ATP and NADPH from ADP, inorganic phosphate (P_i) and $NADP^+$. The light-dependent reactions are summarised in Figure 3.4.6.

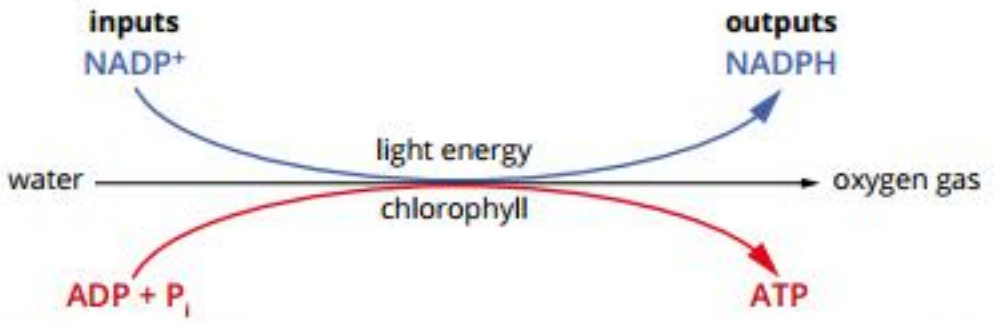


FIGURE 3.4.6 Water is split into oxygen and hydrogen. The oxygen is released as a gas. The hydrogen is added to $NADP^+$. The NADPH and ATP are then used in the second stage of photosynthesis.

LIGHT-INDEPENDENT REACTIONS

The products of the light-dependent reactions, NADPH and ATP, are released into the stroma of the chloroplast. There, they provide energy to drive a biochemical pathway known as the light-independent reactions, sometimes referred to as dark reactions. The light-independent reactions are summarised in Figure 3.4.7.

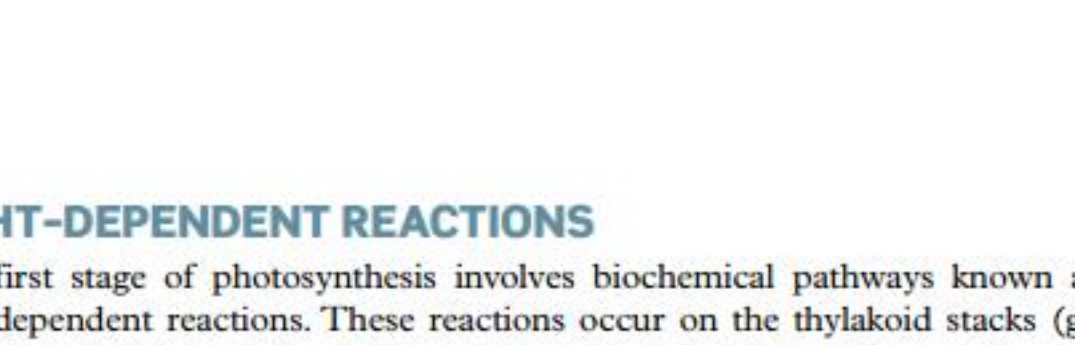


FIGURE 3.4.7 In the second stage of photosynthesis, carbon dioxide is reduced to form the sugar glucose.

The second stage of photosynthesis does not require light for the reaction to be able to proceed. However, it does require the NADPH and ATP of the light-dependent reactions. If NADPH and ATP are present, the reaction can continue in the absence of light (Figure 3.4.8).

The main stage of the light-independent reactions is called the Calvin cycle.

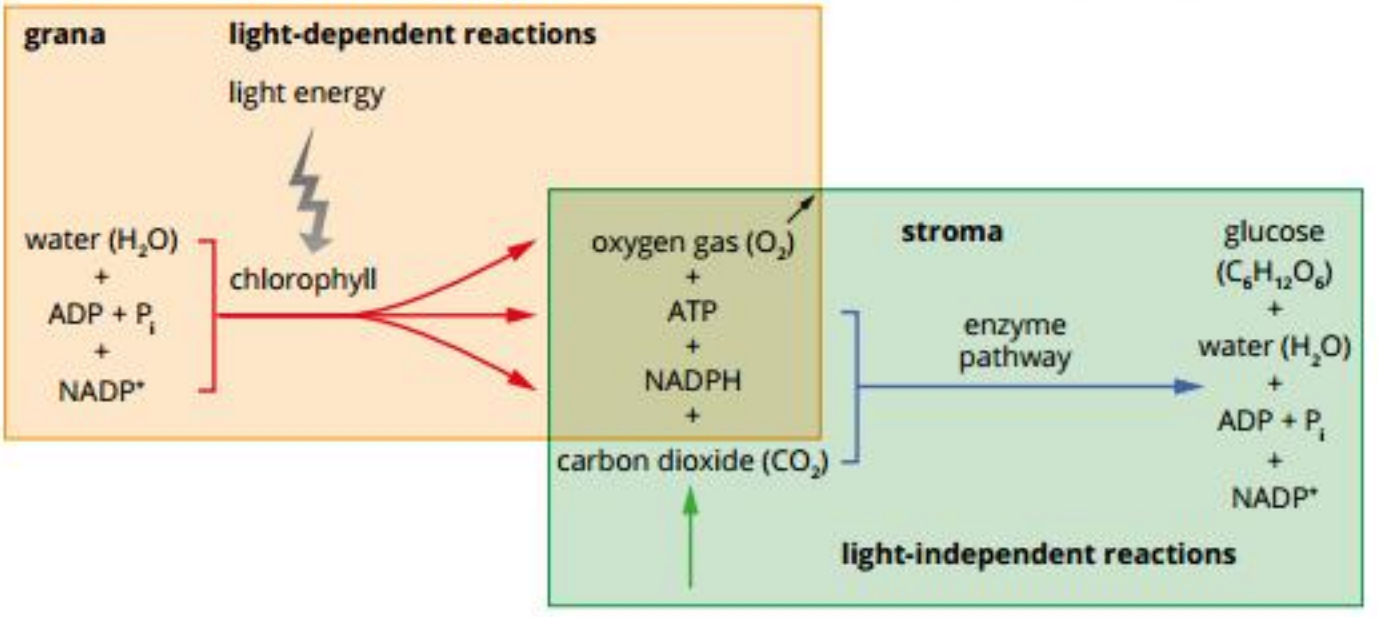


FIGURE 3.4.8 Photosynthesis occurs in two phases. The light-dependent reactions occur in the thylakoid membranes of the grana. The light-independent reactions occur in the stroma and require the carriers generated by the light-dependent reactions.

Calvin cycle

The Calvin cycle comprises all the light-independent reactions. This biochemical pathway of enzyme-catalysed reactions uses carbon dioxide and the energy in the NADPH and ATP produced in the light-dependent reactions to make carbohydrates (Figure 3.4.9). To understand how this process works, it is necessary to realise that each cycle does not exist as a separate entity. There are many Calvin cycles occurring simultaneously in the stroma. If it were possible to isolate a single cycle, then one carbon atom would be added to the system with each 'turn', but no product is actually produced with only one carbon.

The identifiable product of the Calvin cycle is a three-carbon carbohydrate called glyceraldehyde-3-phosphate (GAP). In the cytoplasm, two GAP molecules combine to produce the glucose ($C_6H_{12}O_6$) molecule that is normally identified as the product of photosynthesis. The remaining GAP molecules are recycled into ribulose 1,5-bisphosphate (RuBP), which is the starting molecule of the light-independent reaction.

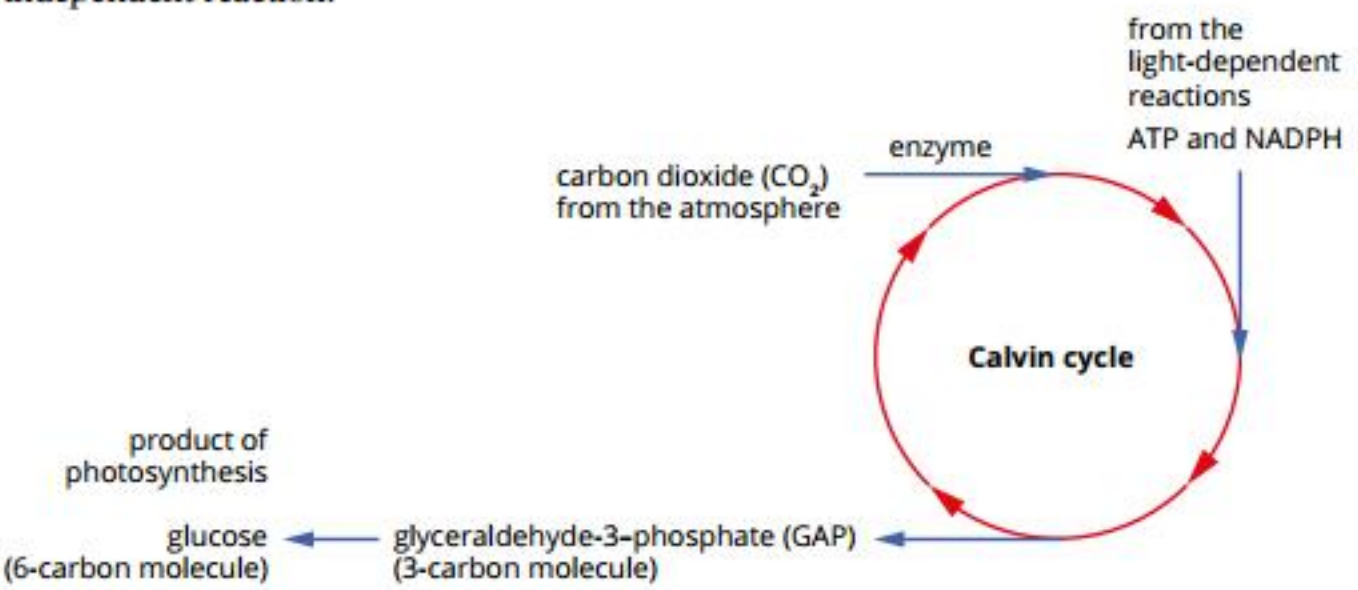


FIGURE 3.4.9 Carbon dioxide from the atmosphere feeds into the Calvin cycle, which uses energy carried by ATP and NADPH from the light-dependent reactions.

FACTORS AFFECTING THE RATE OF PHOTOSYNTHESIS

As for all biochemical processes, the rate of photosynthesis varies according to the internal conditions of the cell, which in turn is often affected by the external environmental conditions. Perhaps most obvious is that the rate of photosynthesis depends on the number of chloroplasts present in a leaf or plant.

Photosynthesis depends on other variables that are interconnected. For example, the opening of **stomata** controls both loss of water and entry of carbon dioxide, as shown in Figure 3.4.10. For this reason, the only way of studying the effect of each factor on the rate of photosynthesis is in the laboratory. Laboratory studies using suspensions of isolated chloroplasts or green algae enable scientists to test the effect of varying the levels of different factors on the rate of photosynthesis under controlled conditions.

The information below outlines the factors that affect the rate of photosynthesis in C_3 plants (3-carbon fixing plants, the most common type of plant). Two other plant types, C_4 and CAM, are discussed at the end of the module.

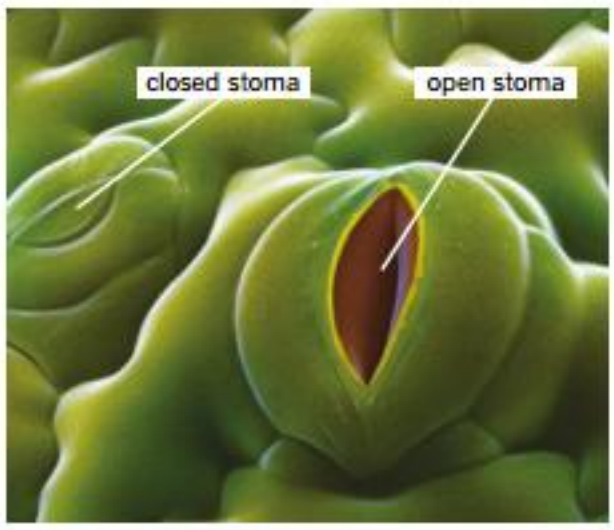


FIGURE 3.4.10 This coloured scanning electron micrograph image of a tobacco leaf shows one open stoma and one closed stoma. When the stoma is open, gas exchange can occur, and water molecules can escape.

Inputs: carbon dioxide and light energy

If you look closely at the process of photosynthesis described in the overall equation, you can predict that the rate at which it occurs will be affected by a number of factors.

$$\text{carbon dioxide} + \text{water} \xrightarrow{\text{light energy}} \text{glucose} + \text{oxygen} + \text{water}$$

$$6CO_2 + 12H_2O \xrightarrow{\text{light energy}} C_6H_{12}O_6 + 6O_2 + 6H_2O$$

FIGURE 3.4.11 This plant has wilted due to water loss. When water is not available, the plant cells lose water, reducing the turgor (pressure) inside the cell. Therefore, the cells cannot maintain a rigid shape, the plant collapses and stomata are closed.

The main requirements of photosynthesis are carbon dioxide, water and light energy. If any one of these factors is in limited supply, it is reasonable to predict that the rate of photosynthesis will be limited also. Because the amount of water used in photosynthesis is small compared with the amount needed to keep the cells alive, a living plant cell normally has sufficient water for photosynthesis to occur. Therefore, water does not have a direct effect on the rate of photosynthesis in nature. However, water does have an indirect effect, because when the plant is suffering from water stress, such as the plant in Figure 3.4.11, the stomata in the leaf close and reduce the availability of carbon dioxide. So, under normal circumstances, photosynthesis is directly affected by the availability of carbon dioxide and light.

Carbon dioxide

The carbon dioxide level in the air remains relatively constant in a natural environment. The factors that affect the CO_2 available for photosynthesis in most terrestrial plants are the number of stomata in the leaves, and whether these stomata are open or closed (Figure 3.4.10). If the stomata are closed, photosynthesis uses up the carbon dioxide inside the leaf, therefore lowering the carbon dioxide concentration in the leaf. With less carbon dioxide available, the rate of photosynthesis, even in the presence of light, is limited.

In the laboratory it is possible to control the concentration of carbon dioxide to which plants are exposed without changing other factors. Figure 3.4.12 shows a comparison of the rate of photosynthesis for a particular species of plant exposed to different concentrations of carbon dioxide at different light intensities.

Terrestrial plants receive their carbon dioxide from the air. Aquatic plants can use the carbon dioxide dissolved in the water.

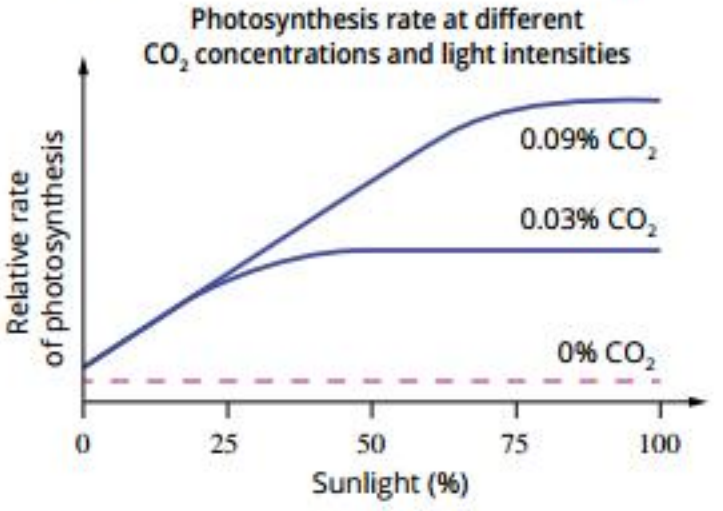


FIGURE 3.4.12 The rate of photosynthesis increases with increasing light intensity at two different concentrations of carbon dioxide compared to no carbon dioxide. At 0.03% carbon dioxide, the rate of photosynthesis increases until approximately 30% sunlight at which point the rate of photosynthesis is limited by the availability of carbon dioxide. At 0.09% carbon dioxide, the rate of photosynthesis continues to increase up to about 80% sunlight. The concentration of carbon dioxide in the atmosphere is approximately 0.04%.

Light

In the laboratory, the chloroplast organelles can be extracted from plant cells and tested in isolation. By varying the amount of light shining on these isolated chloroplasts, while keeping the carbon dioxide and water levels constant, it is possible to measure the rate at which photosynthesis occurs at different amounts of light. The results of this experiment are shown in Figure 3.4.13 and present what is referred to as a light saturation curve. The curve shows a steady increase in the photosynthesis rate with an increase in light intensity until a plateau begins to form. The plateau indicates that there is a maximum rate at which photosynthesis can occur. Assuming unlimited amounts of carbon dioxide (and water), the limit is the point at which all of the photosynthetic systems and enzymes in the chloroplasts are working at their maximum rate.

In the natural environment, the amount of light available to a plant for photosynthesis is determined by the amount of sunlight. Plants in different environments receive different amounts of sunlight, as shown in Figure 3.4.14. The amount of sunlight varies during the cycle of a day, and changes with the seasons and the weather. Figure 3.4.13 shows how tomato plants grown under unshaded conditions have greater rates of photosynthesis than plants grown under shaded (50%) conditions.

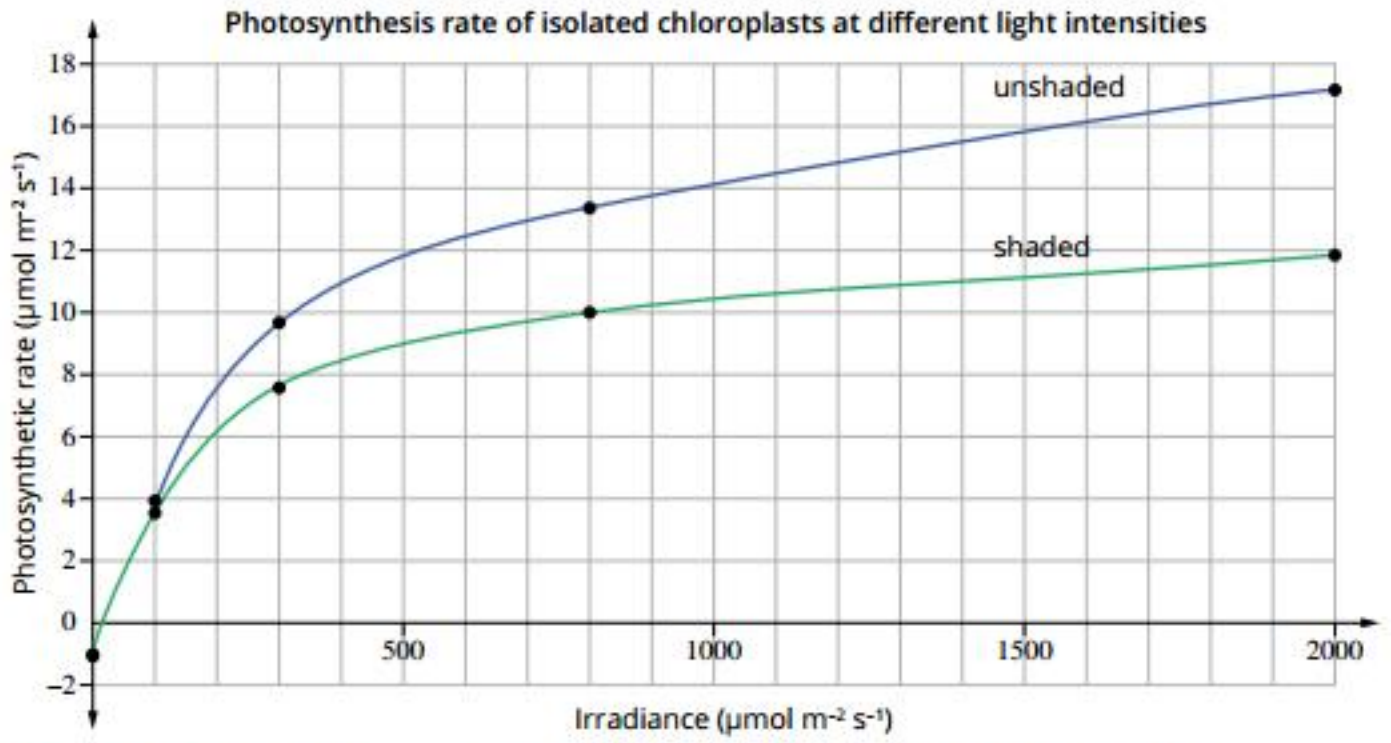


FIGURE 3.4.13 Light saturation curves of chloroplasts extracted from tomato plants grown under shaded (50%) and unshaded conditions

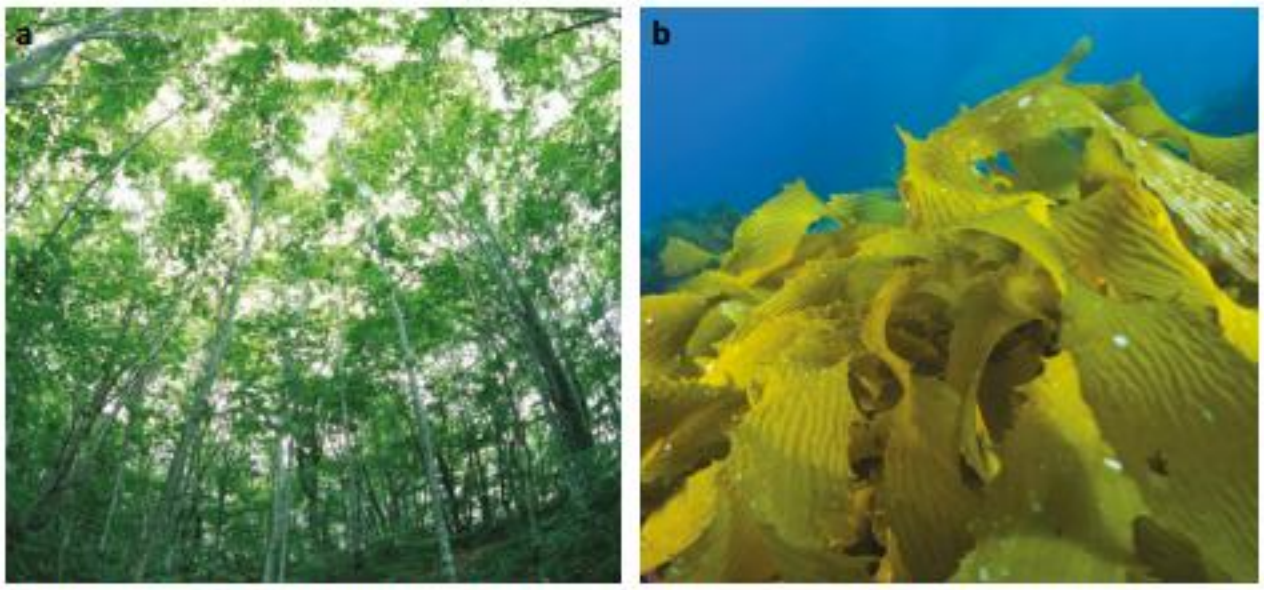


FIGURE 3.4.14 (a) Plants growing on the forest floor receive less light than taller trees. (b) Aquatic photosynthetic organisms, such as this sea kelp, can only grow near the water's surface. As you move deeper underwater, there is less light available.

Temperature

Many enzymes in chloroplasts catalyse the reactions of photosynthesis. Without these enzymes, photosynthesis would not occur. You will recall from Module 3.2 that the rate of an enzyme reaction is affected by temperature and as a consequence the reactions of photosynthesis are affected by temperature. The optimum condition for the functioning of the enzymes of photosynthesis is the temperature at which the maximum rate of photosynthesis occurs. Conversely, at high temperatures, enzymes become denatured and stop functioning, thus ceasing photosynthesis. This is seen in Figure 3.4.15.

Outputs: glucose, oxygen and water

$$\text{carbon dioxide + water} \xrightarrow{\text{light energy}} \text{glucose + oxygen + water}$$

$$6CO_2 + 12H_2O \xrightarrow{\text{light energy}} C_6H_{12}O_6 + 6O_2 + 6H_2O$$

The overall photosynthesis equation shows three basic inputs that produce the three key outputs of glucose, oxygen and water. As will be expected, the amount of products formed from photosynthesis will depend on the input factors outlined above. When inputs are at a high level, the most glucose will be synthesised.

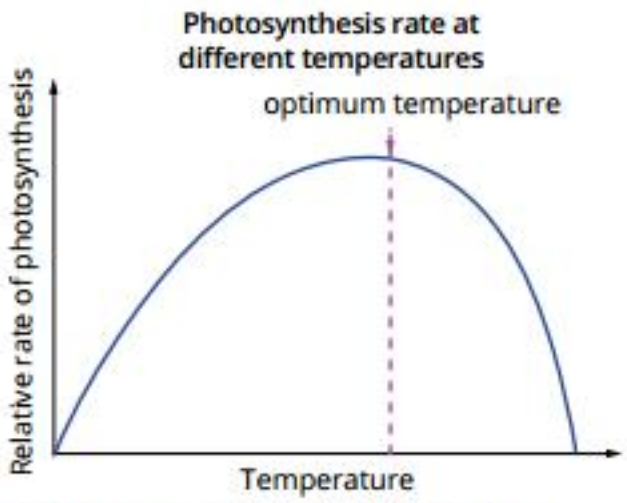


FIGURE 3.4.15 The rate of photosynthesis at different temperatures. The shape of the curve is typical for enzyme reactions. The rate rises with temperature to an optimum temperature and then falls to zero at high temperatures as the photosynthetic enzymes denature.

FIGURE 3.4.16 (a) C_4 (sugar cane) and (b) CAM (pineapple) plants are important commercial crops in tropical Queensland.

The primary output is glucose, a simple monosaccharide carbohydrate. Glucose results from the conversion of solar energy to chemical energy. It contains stored energy that can be used by the plant or consumed by other organisms.

Oxygen is also a product of photosynthesis. Some oxygen will be used in the plant's mitochondria for aerobic respiration and the rest released into the atmosphere from the leaves. The oxygen gas comes from the oxygen atoms of the water, not from the carbon dioxide. Continual output of oxygen by plants into the atmosphere replaces the oxygen used up by animals, as part of the vital carbon dioxide–oxygen cycle.

New water molecules are produced during photosynthesis. Notice that water is both an input and an output for photosynthesis. The six water molecules formed during the reaction are new ones, using oxygen atoms from the carbon dioxide. Researchers discovered this by adding radioactive tags to the input and output molecules when they studied photosynthesis in the laboratory.

Photosynthesis in C_4 and CAM plants

Although photosynthesis seems to be an efficient method of fixing atmospheric CO_2 into a three-carbon molecule, there is a reverse system occurring at the same time. Photorespiration is where the RuBP carboxylase enzyme (which catalyses the key carbon-fixing reaction) also initiates the oxidation of ribulose 1,5-bisphosphate. The result is a loss of between one-quarter and one-half of the fixed carbon that can no longer be synthesised in the Calvin cycle. At higher temperatures, the loss is even greater, posing a distinct problem for the C_3 (three-carbon fixing) plant. This has led to the evolution of two different adaptations—those of the C_4 and the CAM plants.

C_4 plants, which include many grasses and crops, such as sugar cane (Figure 3.4.16a) and sorghum, operate differently in two ways—by fixing a four-carbon molecule rather than the three-carbon molecule made in the Calvin cycle, and by using ATP energy to concentrate CO_2 in mesophyll and bundle sheath cells. The CO_2 binds to the active site on the RuBP carboxylase enzyme, effectively commandeering it for photosynthesis.

CAM plants also fix four-carbon molecules but use crassulacean acid metabolism, or CAM. These plants are generally succulents living in hot climates, and include pineapples (Figure 3.4.16b), cacti and orchids. In CAM plants, the leaf stoma opens at night, not during the day, and so prevent the excess loss of water, and concentrate CO_2 without the use of extra ATP.

3.4 Review

SUMMARY

- Photosynthesis is carried out in two stages: the light-dependent reactions and the light-independent reactions.
- Chloroplasts contain fluid called stroma and thylakoid membranes folded into stacks called grana.
- The light-dependent reactions occur on the grana. Light energy is trapped by chlorophyll, water is split, and oxygen is released as a product.
- The light-independent reactions occur in the stroma. Carbon dioxide is reduced to form glucose.
- The factors affecting the rate of photosynthesis are interconnected.
- An increase in carbon dioxide levels can increase the rate of photosynthesis.
- An increase in light intensity can increase the rate of photosynthesis.
- The availability of chloroplasts to carry out photosynthesis can limit the rate of reaction.
- Plants have an optimum temperature range for photosynthesis because it is controlled by temperature–sensitive enzymes. If it is too cold, the rate of reaction is slow. If it is too hot, the enzymes in chloroplasts can denature.

KEY QUESTIONS

Describe

1 Write the word equation and the chemical equation for photosynthesis.

2 List three things that might happen to the glucose that has been formed by photosynthesis in a plant.

3 State what molecules need to be present for the light-independent phase of photosynthesis to occur.

4 Identify the carbohydrates produced by the Calvin cycle.

5 Create a table listing three factors that directly affect the rate of photosynthesis. Briefly describe how each one does so.

6 Chloroplasts are the site of photosynthesis in a plant cell. Draw a labelled diagram of one chloroplast clearly showing where each of the two stages takes place.

Apply

7 Differentiate the photosynthetic pathways of C_3, C_4 and CAM plants.

8 Using a diagram, represent the two basic phases of photosynthesis and their location in a plant cell.

9 Assess if there is any advantage or disadvantage to plants having a light-independent phase in photosynthesis.

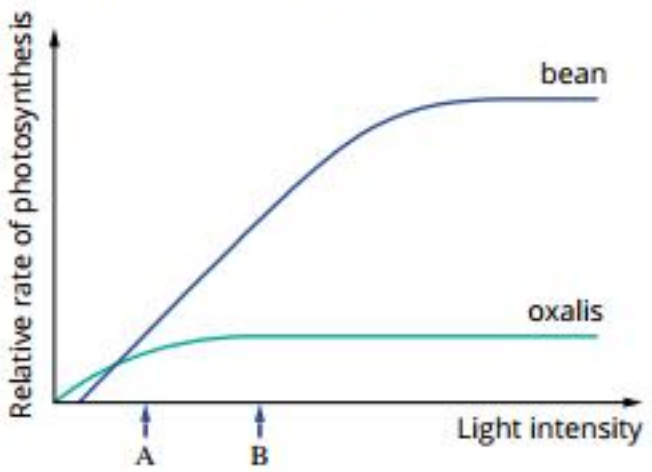
10 The root cells of plants have no exposure to sunlight. How do they obtain energy for growth and survival?

Analyse

11 Using Figure 3.4.4b on page 85, determine the range of wavelengths in which chlorophyll a, b and c will absorb the greatest percentage of light.

12 In a laboratory, radioactive oxygen (^{18}O) was introduced into a closed system with an experimental plant. After a period of exposure to sunlight, the ^{18}O was detected in sugars in the plant's leaf cells. Deduce which molecule with ^{18}O was first introduced into the plant's environment.

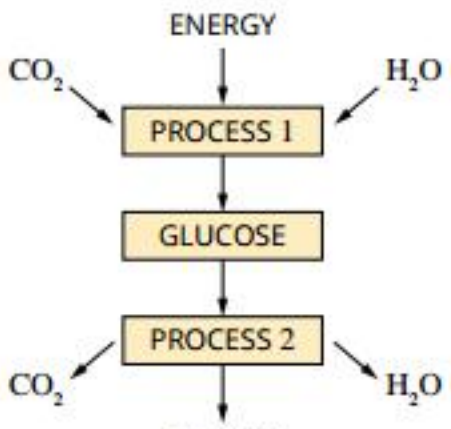
13 The following graph shows the rate of photosynthesis in a bean plant and an oxalis plant.
Draw conclusions with respect to possible limiting factors at points A and B for each plant.



14 The chemical equation for photosynthesis is presented as an overall reaction. Assess the accuracy of this equation and discuss why it is presented this way.

15 A student presented the diagram below to represent energy transfer in plant cells.

- **a** Interpret which processes are represented as 1 and 2.
- **b** Where is each process located in the cell?
- **c** Energy is shown to be both moving in and out in the transfer process.
  - **i** Identify the source of energy for process 1.
  - **ii** Identify the type of energy produced by process 2.
- **d** Write an overall chemical equation for each process.



16 Evaluate the reasons for using isolated cells, mitochondria and chloroplasts in laboratory research to better understand their functions.

Chapter review

03

KEY TERMS

activation energy
active site
adenosine triphosphate (ATP)
aerobic respiration
amino acid
anabolic reaction
anaerobic respiration
autotroph
carbohydrate
carbon dioxide (CO_2)
catabolic reaction
catalyst
cellular respiration
coenzyme
cofactor
cohesive
competitive inhibition
denature (or denaturation)
electron transfer chain
enzyme
enzyme–substrate complex
$FADH_2$ (flavin adenine dinucleotide)
fermentation
glucose
glycolysis
heterotroph
Krebs cycle
lactic acid
lipid
NADH (nicotinamide adenine dinucleotide)
oxygen (O_2)
photosynthesis
protein
proteomics
pyruvate
stomata (sing. stoma)
substrate

KEY QUESTIONS

Describe

1 Identify which of the following lists represents key organic compounds in cells.
- **A** carbohydrates, water, proteins lipids
- **B** water, carbon dioxide, oxygen, enzymes
- **C** carbohydrates, proteins, lipids, nucleic acids
- **D** carbohydrates, proteins, lipids, minerals

2 Identify which of the following is not made of monomers and does not form a polymer.
- **A** carbohydrate
- **B** lipid
- **C** nucleic acid
- **D** protein

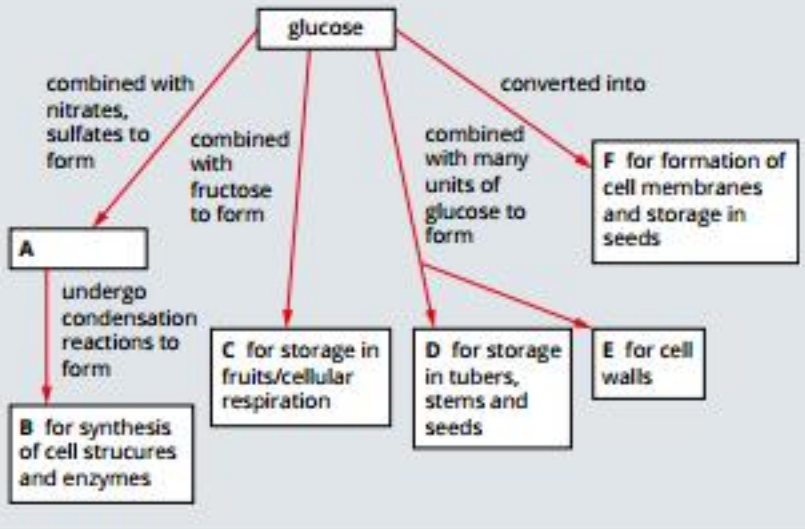
3 An organic molecule required to speed up the rate of a biochemical reaction is:
- **A** a reactant
- **B** an enzyme
- **C** an enzyme activator
- **D** a vitamin

4 Recall that enzymes reduce the activation energy of a reaction by:
- **A** bringing the reactants close together so that a reaction is more likely to occur
- **B** orientating the reactants in the most favourable position for the reaction
- **C** providing a micro-environment favourable to the chemical reaction
- **D** all of the above

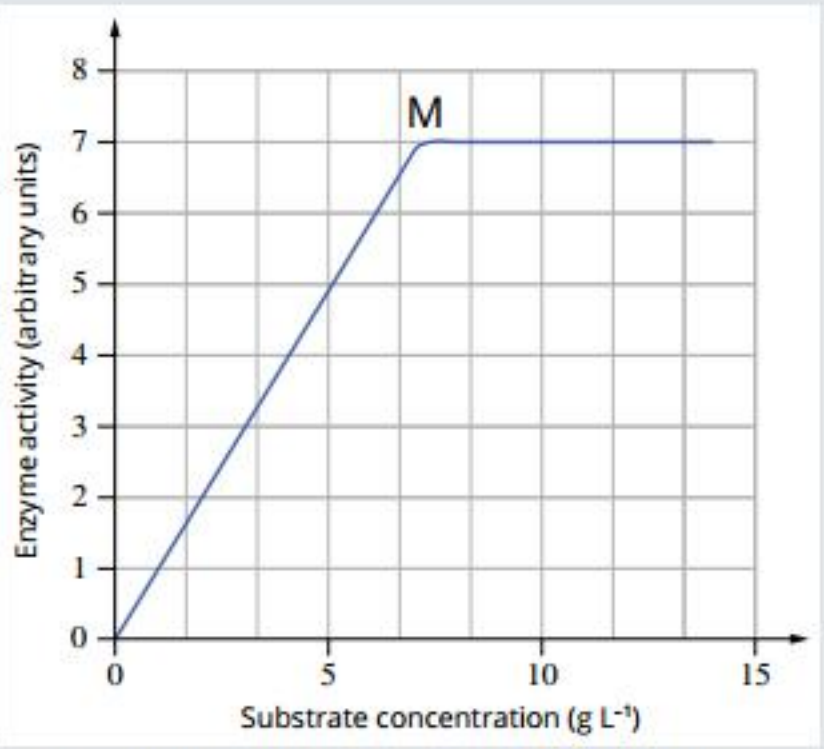
5 When a metabolic process breaks down large molecules into smaller ones, it is said to be exergonic because it releases energy. The process is also said to be:
- **A** endergonic
- **B** catabolic
- **C** anabolic
- **D** catalytic

6 Identify which of the statements about NAD^+ is not true.
- **A** NAD^+ can accept a hydrogen ion.
- **B** When NADH gives up a proton, NAD^+ results.
- **C** Only a small amount of NAD^+ is needed in a cell.
- **D** NAD^+ can only be used once before it must be resynthesised.

7 The diagram is a summary of the major processes in plants that occur after photosynthesis. Identify the correct term for each of the letters A–F. For B–F you are required to replace each description with the relevant single word.


glucose
combined with nitrates, sulfates to form
combined with fructose to form
combined with many units of glucose to form
converted into
F for formation of cell membranes and storage in seeds
A
undergo condensation reactions to form
C for storage in fruits/cellular respiration
D for storage in tubers, stems and seeds
E for cell walls
B for synthesis of cell strucures and enzymes

8 A student investigating the activity of the enzyme pepsin, which is found in the stomach of humans, observed the change in enzyme activity as the concentration of the substrate (protein) increased. The experiment was conducted at pH 3 and 37°C. The student's data was presented in the graph shown.



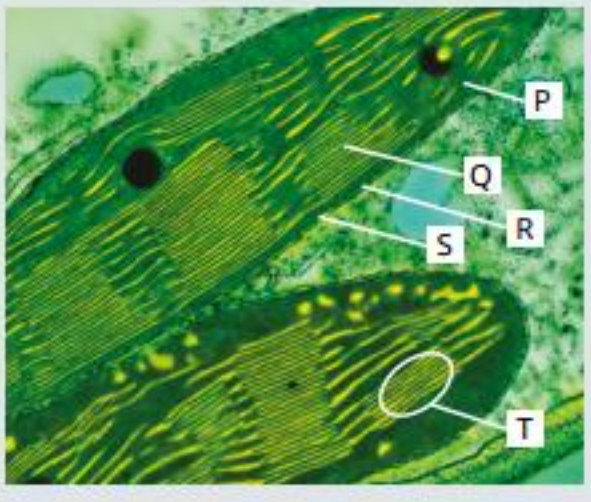
The student wanted to change the experiment so that the rate of reaction at point M was higher than that shown. In order to do this the student could:

A increase the pH to 8
B decrease the temperature
C increase the amount of enzyme
D increase the concentration of the substrate

9 Identify whether each of the following statements is true or false.

a Chlorophyll is a pigment that causes green colouring of leaves.
b Chloroplasts are pigments that absorb light.
c Chlorophyll is located in the thylakoid membranes.
d Chlorophyll absorbs light energy, which is then transformed into chemical energy.

10 The figure below shows a transmission electron micrograph of a chloroplast. Identify structures P–T.



11 Which of the following is true regarding glycolysis?

A It produces 36 ATP.
B It is an aerobic process.
C It uses glucose as a reactant.
D It occurs in the mitochondria.

12 Chemical compounds can be classified as organic or inorganic.

a Define 'organic compound'.
b Create a table to answer parts i and ii.
i Identify each compound in the list below as an organic or inorganic compound: carbon dioxide, maltose, maltase, cellulose, cholesterol, Vitamin D, sodium nitrate, calcium, insulin, oestrogen
ii For the organic compounds, classify them further as one of the four types of organic molecules.

13 Briefly describe what happens to pyruvate in the two processes that can occur after glycolysis when oxygen is:

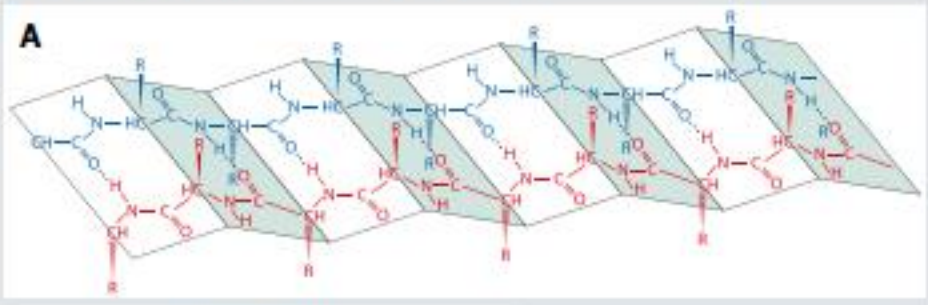
a present
b absent

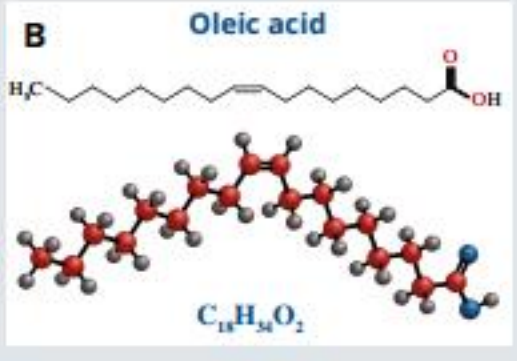
Apply

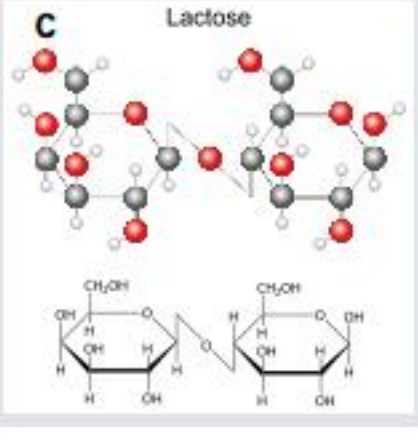
14 Explain why photosynthesis is not the opposite of aerobic respiration.

15 The three diagrams below are models that represent three different types of biomolecules.

a Identify which group of compounds each belongs to.
b Explain your answer for each.







16 Do both plants and animals require minerals? Give examples to justify your answer.

17 The main function of cellular respiration is to:

A use the energy stored in food to generate ATP for cellular work

B use sunlight to produce sugar

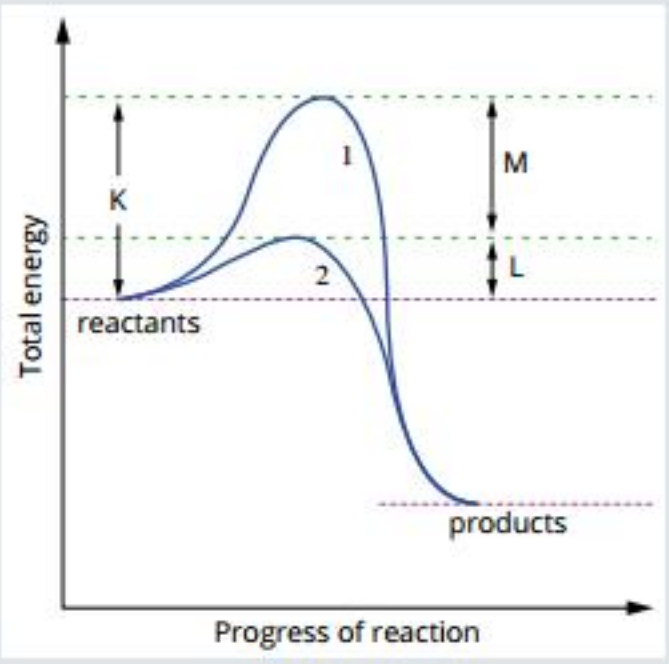
C get rid of the carbon dioxide that builds up in working cells

D break ATP down to ADP so that cells can perform work.

18 Explain why an enzyme functions effectively even when its concentration is low.

19 Deduce why there are thousands of different enzymes in the human body.

20 The breaking or formation of bonds between atoms during biochemical reactions results in changes in the energy content of the molecules. Formation of new chemical bonds requires energy and the breaking of chemical bonds releases energy. The graph shows the energy changes during one particular chemical reaction with and without an enzyme.



a Describe the energy change that is represented by:

i K

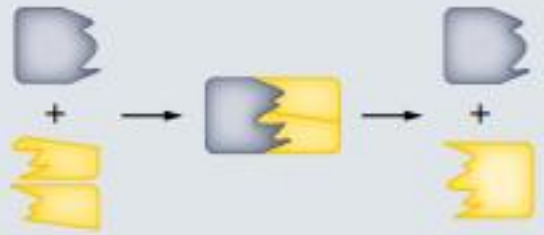
ii L

iii M

b Determine if the overall reaction is exergonic (releases energy) or endergonic (requires energy).

c Explain which line shows the enzyme-catalysed reaction.

21 The following diagram illustrates one model of enzyme activity.



a Explain which model of enzyme activity is shown in this diagram.

b The structures of both substrates in the diagram have changed slightly. Explain why this would, or would not, be an indication that the enzyme could act on a range of substrates.

22 Many chemical reactions in living things occur as part of a metabolic pathway. Metabolic pathways involve a series of reactions with a different enzyme catalysing each step in the pathway. In these pathways, the product of one reaction is the substrate for the next reaction. The following diagram shows the enzymes involved in one such pathway.

The enzyme substrates are shown below.

a **i** Match each substrate with its enzyme.

ii Explain the basis of your decision.

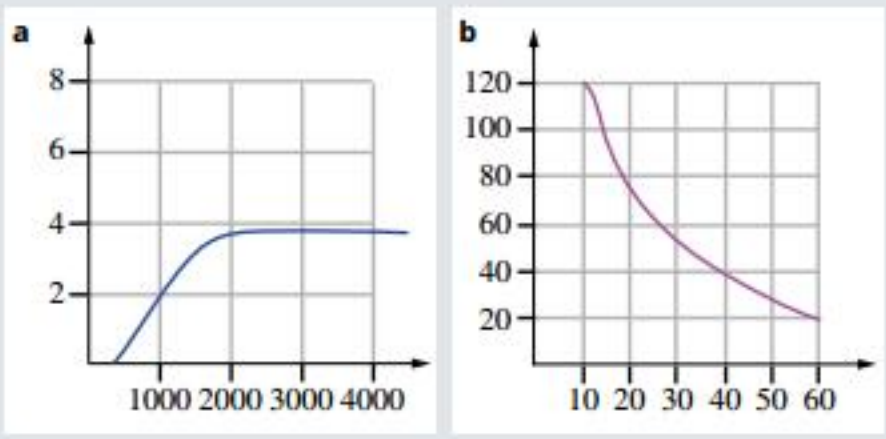
b The following molecule is the final product of the reaction.

i If the concentration of the final product builds up in the cell, the reaction stops. Explain how the increase in the concentration of the final product stops the reaction proceeding.

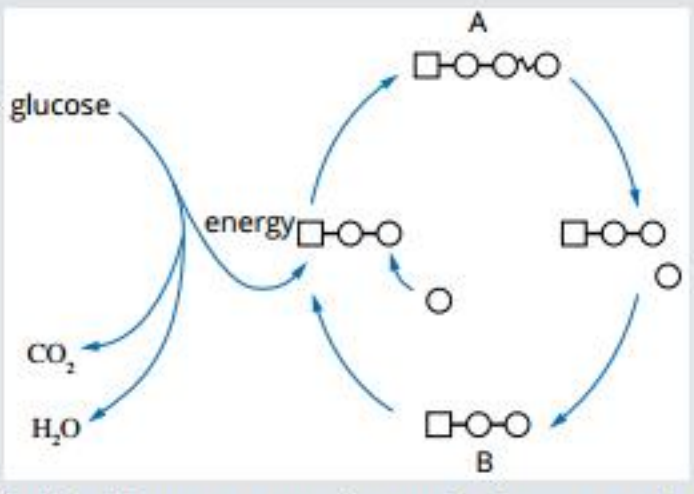
ii Determine whether the product is acting as a competitive inhibitor. Explain your answer.

23 There are two stages of photosynthesis. Summarise what process occurs in each stage.

24 The following graphs represent the changes in rate of photosynthesis when temperature, light intensity or distance from a light source are increased. Label each graph with the factor that is best represented by the data presented.



25 Study the diagram shown, which summarises the processes of energy release in cells.



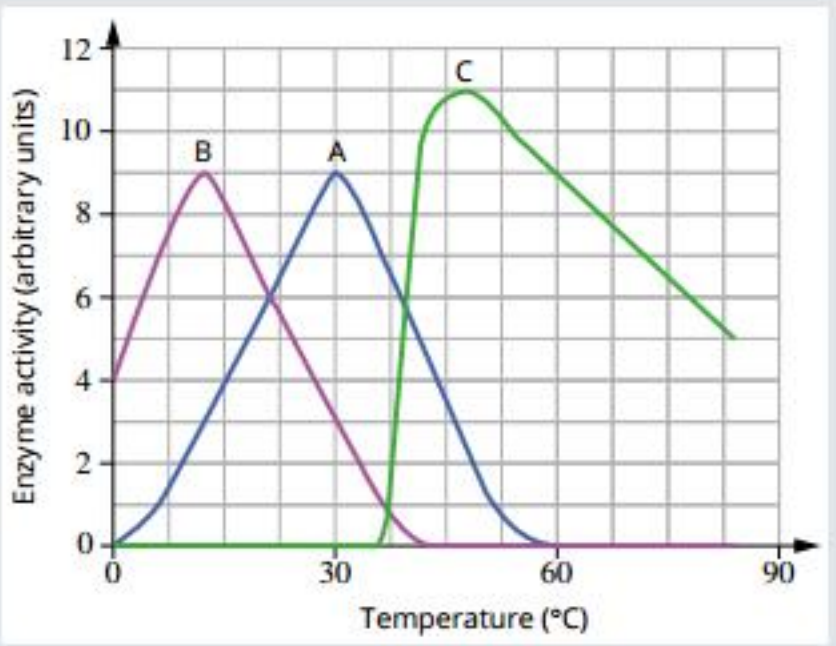
a In the diagram, name the molecule represented by the letter:
- **i** A
- **ii** B

b Describe how molecules A and B are related.

c Glucose is the energy-rich molecule that enters the glycolysis process. Describe the products of this process for each molecule of glucose.

Analyse

26 An experiment was performed to investigate enzyme activity in three different species: the two-toed sloth (a mammal), an Arctic trout, and a bacterium from a thermal spring. The activities of the enzymes from each organism are plotted on the graph below.



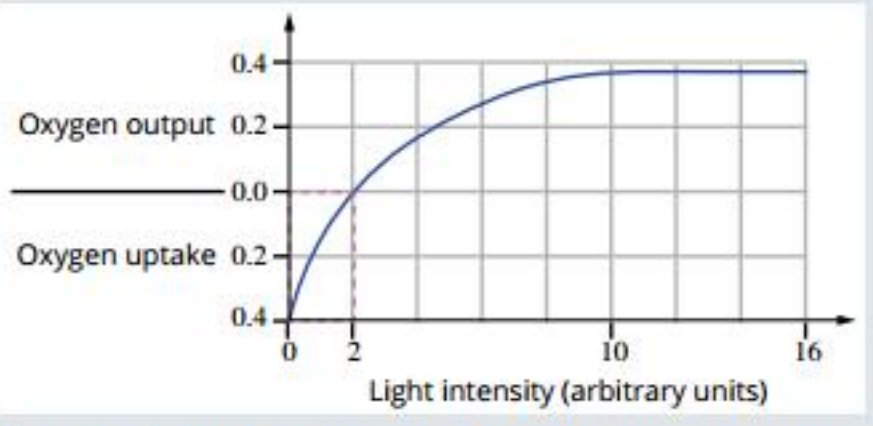
a Explain which graph belongs to each animal.

b Explain why no activity was observed at 60°C for enzyme A.

27 The following graph shows the relationship between net oxygen uptake or output and light intensity for a green plant. Explain what is happening when light intensity is at:

a 2 units.

b 16 units.



28 **a** Describe the energy-releasing processes that occur in the leg muscles of a sprinter in a 50 m race.

b Explain how the muscles recover their normal states after the race is over.

c Describe how these processes would be different in the leg muscles of someone who went for a gentle jog.

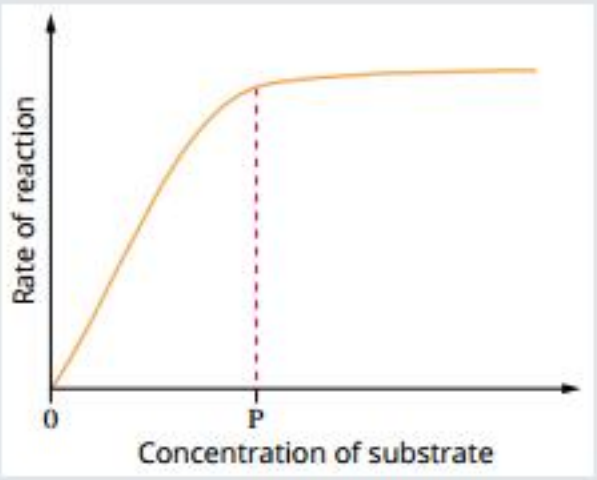
d Propose why the muscles of a sprinter might be sore 2 days after the race.

29 Pepsin is an enzyme that is released into the stomach of humans (pH 1.5–3.5, 37°C), where it breaks down proteins into polypeptides. Predict how the activity of pepsin would change as the:

a temperature is increased from 37°C to 45°C.

b pH is increased above 5.

30 The following graph illustrates the relationship between the concentration of an enzyme substrate and rate of reaction.

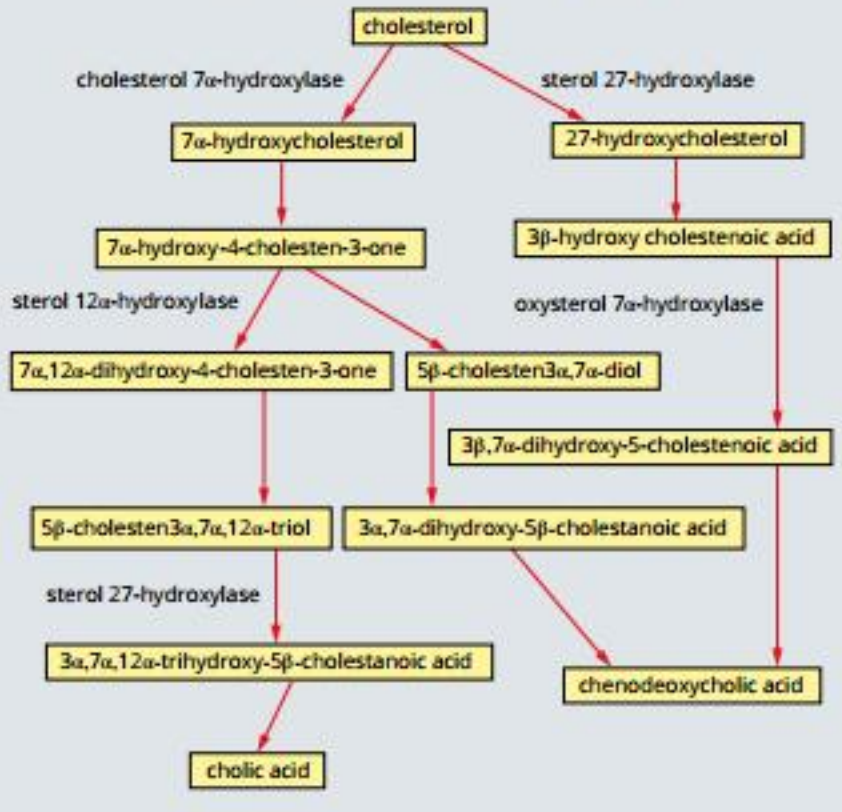


a Determine the relationship between the substrate concentration and the rate of reaction from 0 to P units of substrate concentration.

b Infer what will happen at and after point P.

c Explain what it means when the concentration of the enzyme is described as being a limiting factor.

31 Bile acids are processed in the liver to form bile salts. Bile salts are used in the small intestine to mechanically break down fats in the diet.
The metabolic pathway for the formation of two bile acids is shown in the flow chart. The chart is simplified; some steps in the pathway between 3b,7a-dihydroxy-5-cholestenoic acid and chenodeoxycholic acid are not known, nor are some of the enzymes in the pathway.



a One enzyme, used twice in the pathway, is sterol 27-hydroxylase. If an individual was unable to make functional sterol 27-hydroxylase, conclude whether they would be able to make either of the bile acids shown in the flow chart.

b Determine whether it is likely that chenodeoxycholic acid acts as an inhibitor of sterol 27-hydroxylase.

c Infer how sterol 27-hydoxylase can catalyse two different steps in the pathway.

32 During chemical reactions, changes in free energy occur. The change of energy is given as ΔG (delta G). A positive energy change means that there had to be an input of energy for the reaction to occur (endergonic) and a negative energy change means the reaction released energy (exergonic). Energy released by one step can be used in subsequent steps.
A particular metabolic pathway involves four enzymes (K, L, M and N) in a four-step pathway. Molecule A is the initial substrate of the pathway and molecule E is the final product.

$$A \xrightarrow{K} B \xrightarrow{L} C \xrightarrow{M} D \xrightarrow{N} E$$

The energy requirements of each step are shown in the table below.

Step	ΔG (joules)	Enzyme
A → B	+5	K
B → C	+2	L
C → D	−6	M
D → E	−4	N

a Determine the exergonic steps.

b Assess whether the pathway is anabolic or catabolic.

c The shape of enzyme K is shown below. High concentrations of molecule E inhibit enzyme K.

i Deduce the shape of the substrate for enzyme K.

ii Draw a possible shape for molecule E.

iii Determine, using annotated diagrams, how molecule E inhibits enzyme K catalysing the formation of molecule B, thereby regulating the pathway.

33 Atrazine is a chemical commonly used as a weed killer. It is absorbed by roots from the soil. In the leaves, it attaches to a protein, called D1, which is a part of the electron transfer chain used to generate ATP during photosynthesis. It blocks the movement of electrons along this chain.

- **a** Deduce which phase of photosynthesis is disrupted by atrazine.
- **b** ATP is not a final product of photosynthesis. Determine what happens to the ATP normally produced in the electron transfer chain.
- **c** The advertising blurb on one particular brand of weed killer containing atrazine as its active ingredient says, '... this product works by starving the plant ...'. Evaluate this description of the action of atrazine.

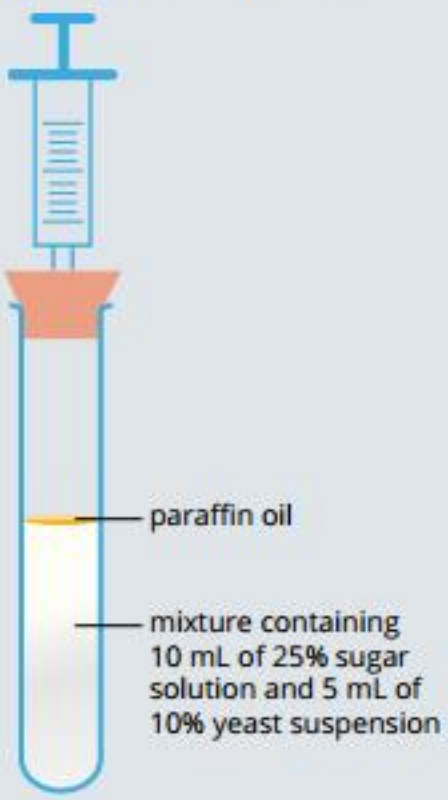
34 A student investigated the effect of varying types of sugar solutions (glucose, lactose, fructose, maltose and sucrose) on the rate of anaerobic respiration in yeast. The student hypothesised that the rate of respiration in yeast will be the highest for glucose. The steps in the experiment are described below.

Step 1: 10 mL of fructose solution (25% concentration), 5 mL of yeast suspension (10% concentration) and three drops of paraffin oil were added to a test-tube and mixed thoroughly.

Step 2: The test-tube was sealed with a rubber stopper with a syringe attached, as shown in the diagram.

Step 3: After 15 minutes, the volume of gas collected in the syringe was recorded.

Step 4: Steps 1 to 3 were repeated four more times. The experiment was then repeated using four other sugars: glucose, lactose, maltose and sucrose. The results are shown in the table below.



Volume of gas collected (mL)						
sugar	set 1	set 2	set 3	set 4	set 5	mean
fructose	1.4	1.4	2.0	2.6	5.4	
glucose	9.0	8.2	7.0	8.6	8.8	
lactose	0.0	0.0	0.0	0.0	0.0	
maltose	2.8	3.2	3.0	3.0	6.2	
sucrose	7.4	7.0	7.0	6.6	6.8	

- **a** Define 'anaerobic respiration'.
- **b** Write the word equation that represents the anaerobic respiration of yeast.
- **c** Determine the purpose of the paraffin oil in this experiment.
- **d** Calculate the mean for the experimental results to complete the table. Use the calculated means to plot a graph of the data. Ensure the graph is labelled appropriately.
- **e** Assess which type of sugar resulted in the:
  - **i** highest rate of respiration
  - **ii** lowest rate of respiration
- **f** Judge whether or not the student's hypothesis was supported by the experimental results.
- **g** Propose a suitable control for this experiment.

Interpret

35 Many junior science books state that 'All enzymes are proteins. Each enzyme is specific to one reaction'. Appraise the truth, or otherwise, of this statement.

36 Imagine you are a scientist who has published a textbook for students in which the lock-and-key mechanism has been explained in detail. Discuss how you would react to a new model being proposed with scientific evidence to support it.

37 Many people believe that plants convert carbon dioxide into oxygen. Explore this belief.

38 **a** Explain why the rate of photosynthesis for a plant might decrease under hot and windy conditions.

- **b** Design a scientific experiment to test for such changes.

Data analysis

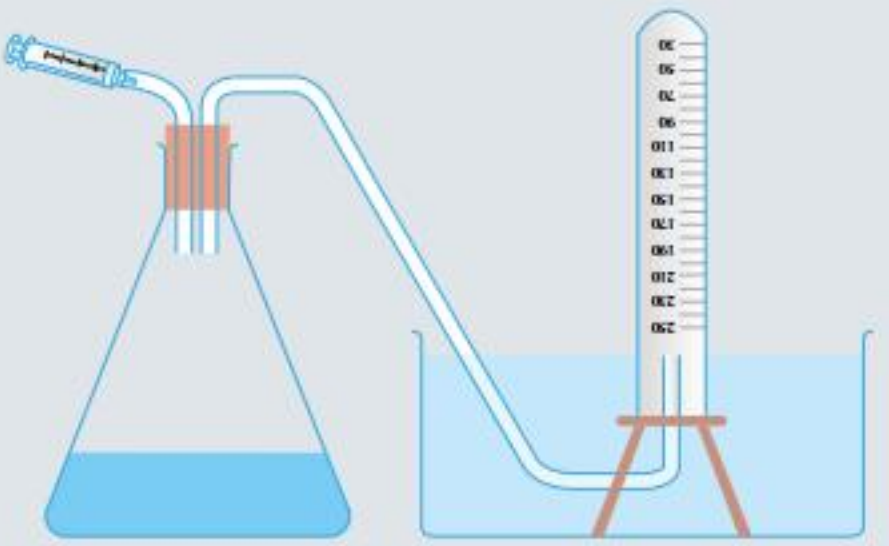
DATA SET 1

The information below applies to Questions 1–5.

Cellular respiration in the mitochondria produces a toxic by-product called hydrogen peroxide (H_2O_2). Even though H_2O_2 spontaneously breaks down to form water and oxygen, all living things contain an enzyme called catalase, which speeds up the detoxification reaction.

The reaction is: $2H_2O_2 \rightarrow 2H_2O + O_2$

A group of students performed an experiment to investigate the effects of varying substrate concentration on the activity of catalase at air temperature. Potatoes were blended into a paste and strained to collect potato juice, which contains the enzyme catalase. The experimental setup below shows how oxygen gas, from the reaction in the flask, was collected by displacement of water in a measuring cylinder. 20 mL of potato juice was placed in the flask, the flask was sealed, and a syringe was used to add hydrogen peroxide.



In the first test, 2 mL of a 5% solution of H_2O_2 was used. Subsequent tests used 2 mL of 10%, 15%, 20% and 25% solutions of H_2O_2. The volume of oxygen produced in 5 minutes was measured. The results obtained by the students are shown in Table 1.

TABLE 1 Catalase reaction and oxygen production

Concentration of H_2O_2 (%) (independent variable)	Volume of oxygen collected (cm^3) (dependent variable)
5	9.5
10	20
15	31
20	40.5
25	52

Question 1 (1 mark)

Explain why the volume of oxygen produced can be used as a measure of the activity of catalase.

Question 2 (4 marks)

a Plot a graph of the students' results.

b Describe the relationship between the volume of oxygen produced and the concentration of H_2O_2.

c Would it be accurate to plot a 0,0 point on the graph? Justify your answer.

Question 3 (2 marks)

a Write a suitable hypothesis for this experiment.

b Draw a conclusion from the data.

Question 4 (1 mark)

Predict the volume of oxygen produced if 2 mL of 30% solution of H_2O_2 was used.

Question 5 (2 marks)

Infer the effect on catalase activity in the same experimental setup if the temperature of reactants in the flask is varied as below:

a the flask stands in an ice bath at about 4°C.

b the flask stands in hot water bath at about 50°C.

CHAPTER

04 Cell organisation and specialisation

In this chapter, you will learn about the organisation of cells in complex multicellular organisms, and how cells become specialised. As multicellular organisms increase in complexity, their cells become more highly organised. The levels of organisation in multicellular organisms are specialised cells, tissues, organs and systems. You will look at each of these levels of organisation and the specialised structures and functions that have evolved to meet the needs of complex organisms.

Finally, you will learn about the significance of stem cells in the development and maintenance of complex multicellular organisms and the role of stem cells in cell specialisation.

Syllabus subject matter

Topic 1 • Cells as the basis of life

- Describe how the hierarchical organisation of cells, tissues, organs and systems allow multicellular organisms to
  - obtain nutrients, e.g. digestive and circulatory systems
  - exchange gases, e.g. respiratory and circulatory systems
  - remove wastes, e.g. respiratory, circulatory and excretory systems. **4.1***
- Explain that each body system contains specialised cells and tissues that are structurally suited to function, including
  - size and shape (SA:V ratio)
  - organelle composition. **4.1***
- Describe how stem cells originate through the process of mitosis and differentiate into specialised cells to form tissues. **4.2**
- Distinguish between unipotent, multipotent, pluripotent and totipotent stem cells. **4.2**

*The greyed-out dot points are also addressed in Chapter 5.

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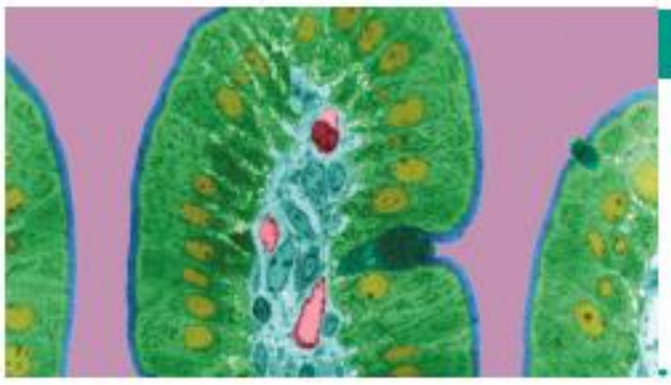
4.1 Levels of organisation in multicellular organisms



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand the levels of cellular organisation in multicellular organisms
- be able to identify specialised cells, tissues, organs and systems
- be able to relate a specialised cell's structure to its function
- understand the levels of cellular organisation in complex plants and animals.

FIGURE 4.1.1 A complex multicellular organism, such as a koala, is more than just a mass of cells. The cells are organised so that they can work together to carry out the functions required for life.

Cells carry out all of the functions necessary to sustain the life of an **organism**, including obtaining nutrients and water, exchanging gases, sourcing energy, removing waste products and reproducing. As you will recall from Chapter 2, **unicellular** organisms consist of a single cell and **multicellular** organisms consist of many cells. In unicellular organisms, all the functions required to survive and reproduce are carried out by just one cell. In multicellular organisms, many cells work cooperatively for the survival and reproduction of the organism.

In this module, you will look more closely at multicellular organisms and how they are organised to overcome challenges and take full advantage of multicellularity.

MULTICELLULARITY

All multicellular organisms consist of eukaryotic cells. There is an enormous diversity of multicellular organisms, from simple mosses, sponges and corals to complex flowering plants, birds and mammals. A multicellular organism can consist of many trillions of cells. In a mammal, such as the koala in Figure 4.1.1 or a human, these cells consist of hundreds of different types of specialised cells. In mammals, examples of specialised cells include muscle cells, red blood cells, bone cells and nerve cells. Each of these cell types has a specific function that contributes to the survival and reproduction of the whole organism.

Although multicellularity has many advantages, it also has a number of challenges. An individual muscle cell is capable of shortening, yet on its own could not possibly bring about movement in a large organism. This requires the aid of numerous other muscle cells that have differentiated and specialised. The muscle cells must also work with other types of cells, such as specialised cells in bones and tendons. Other specialised cells are needed to bring nutrients to the muscle cell and to get rid of waste products. To ensure the cells can carry out their functions correctly and maintain healthy systems, the organism must expend energy finding the resources it needs to fuel all of its cells.

For an organism to be considered truly multicellular:

- its cells must have the same DNA (except the reproductive cells)
- its cells must be connected and must communicate and cooperate to function as a single organism
- it must have different cells that are specialised and responsible for specific functions (including reproduction)
- its cells must be dependent on each other for survival.

LEVELS OF CELLULAR ORGANISATION

A cell must be able to obtain nutrients and remove waste, and physical conditions such as temperature, solute concentration and pH must remain within the tolerable limits of the cell. If any of these conditions are not met, the cell dies. This is also true for the individual cells that comprise a multicellular organism.

One advantage of being a large multicellular organism is that most body cells are isolated from the external environment by a protective outer layer called the **epidermis**. This outer layer provides a buffer against changes in the external environment, allowing conditions on the inside of the organism to be maintained at suitable levels for the efficient functioning of cells. For example, your epidermis (skin) operates as a barrier to keep your internal environment separate from the external environment (Figure 4.1.2).

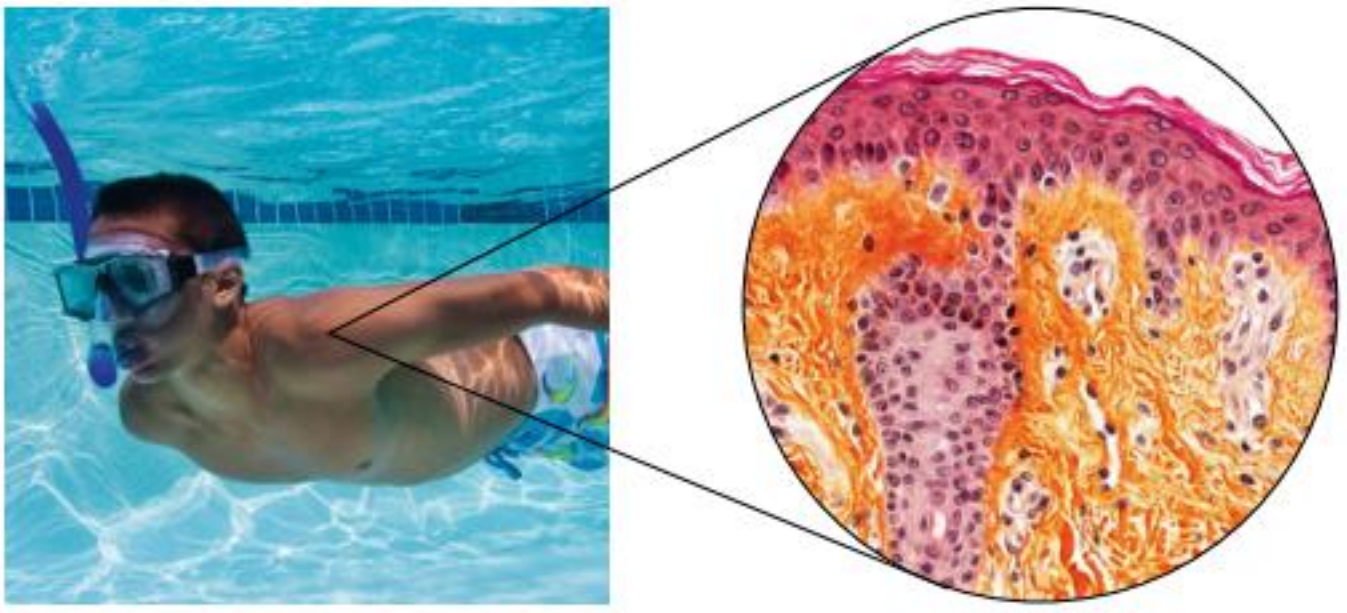


FIGURE 4.1.2 The epidermis provides a barrier between the internal and external environment of multicellular organisms, which helps to maintain the stable internal environment of multicellular organisms. The epidermis comprises several layers of specialised cells. Only the uppermost layer (pink) is in contact with the external environment.

However, the isolation of the internal environment from the external environment means that most cells do not have direct access to their essential requirements, such as oxygen. It also means that wastes expelled from the cells need to be removed from the internal environment so that they do not accumulate around the cell. As an organism increases in size and complexity, greater cooperation and coordination is required between its cells—the cells must be organised.

Multicellular organisms, depending on their complexity, can be organised into the following levels to provide the needs of the entire organism:

- specialised cells
- tissues
- organs
- systems.

Specialised cells

Specialised cells are cells that have a specific function. All cells perform particular jobs in a multicellular organism and have unique structural adaptations that enable them to carry out these functions. Some specialised cells move independently around the body, such as red blood cells. However, most specialised cells form the building blocks of complex tissues and **organs** in multicellular organisms. Examples of specialised cells in plants are root hairs, palisade cells and guard cells. In animals, specialised cells include myocytes (muscle cells), erythrocytes (red blood cells), epithelial cells and neurons. Figure 4.1.3 shows some neurons.

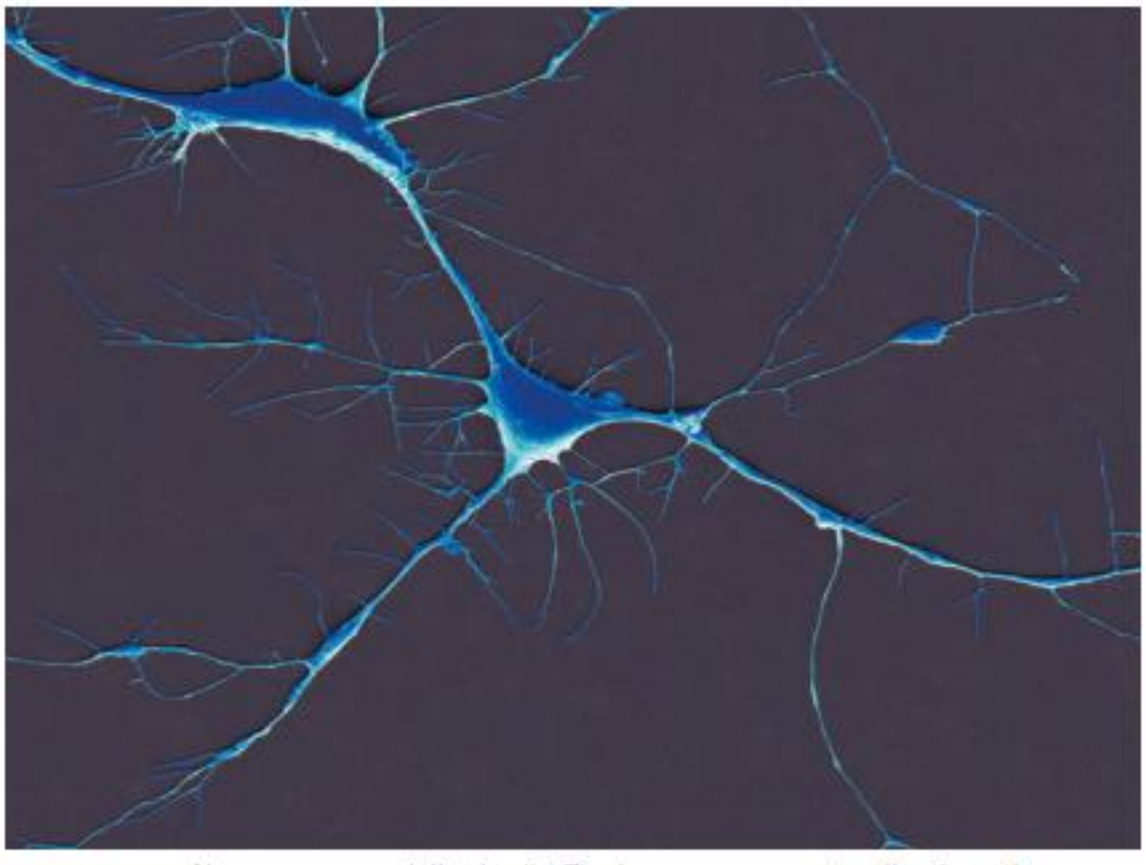


FIGURE 4.1.3 Neurons are specialised cells. The long processes extending from the cell body transmit nerve impulses away from the neuron.

The term 'organism' is derived from the French word *organisme*, which means 'organise'.

Tissues

Specialised cells are organised into **tissues**. Tissues are groups of similar cells working together to carry out a particular function in a multicellular organism. For some organisms, this level of organisation is sufficient to meet all its needs.

But as organisms become more complex, tissues alone may not be enough to carry out all the tasks required, and it is reasoned that tissues have evolved to group together in distinct structures called organs.

Organs

An **organ** consists of two or more tissues that work together to perform one or more specialised tasks. An organ is commonly recognisable as a distinct structure. Examples of organs are flowers, leaves and roots in vascular plants, and heart, liver and brain in mammals.

Systems

In multicellular organisms, an organ rarely functions independently of other organs. Instead, organs form an organ system, commonly referred to simply as a system. A system is a group of organs that work together to perform a vital task, such as the circulatory and respiratory systems in humans.

Organisms

The final level of organisation is the organism itself. In a complex animal, systems work together and contribute to the successful functioning and reproduction of the whole organism. Figure 4.1.4 shows the levels of organisation in animals and plants.

i As biological structures and functions become more complex, cells become more and more specialised.

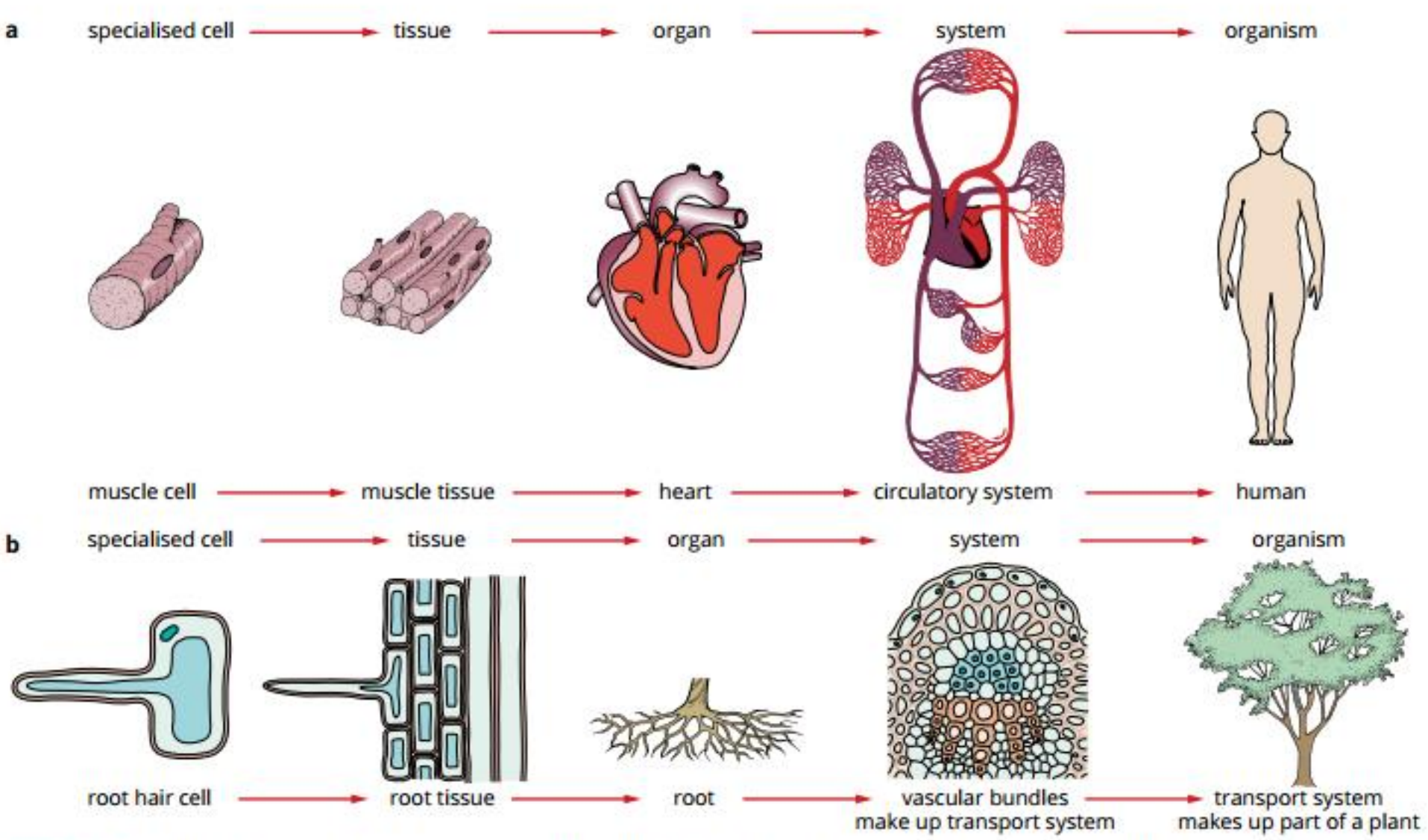


FIGURE 4.1.4 (a) The levels of organisation in a complex multicellular animal: cell, tissue, organ, system and organism. (b) The levels of organisation in a complex multicellular plant: cell, tissue, organ, system and organism

ORGANISATION IN SIMPLE MULTICELLULAR ORGANISMS

Some multicellular organisms are organised only at the cellular level. This includes simple multicellular organisms such as sponges and sea jellies. These animals are considered to be tissueless multicellular organisms because their cells are not organised into discrete, functioning systems within the organism. Nevertheless, the cells in these simpler multicellular organisms are still organised to enable the organisms to obtain nutrients, exchange gases, and remove wastes.

Although simple multicellular organisms are more complex than unicellular organisms such as *Euglena*, simple multicellular organisms can survive without organising their cells into true tissues and organs because they are often only a few cells thick. This means that materials can diffuse easily into and between cells. This lack of organisational complexity also means that many simple multicellular organisms, such as planarians, can regenerate, building new limbs or even an entirely new organism from just a tiny piece of their body or a single cell (see Module 4.2).

In sponges, such as the azure vase sponge in Figure 4.1.5, the body is hollow and consists of two layers of eukaryotic cells separated by a jelly-like substance. The outer layer protects the sponge and also contains tiny pores through which water and food can enter. Sponges are filter feeders; they filter plankton, bacteria, dinoflagellates and many other microscopic organisms from the water around them. Digestion is carried out within food vacuoles inside the cells of the sponge. The inner layer consists of a number of cell types, including collar cells and amoebocytes. The organisation and coordination between these layers of cells enables the sponges to obtain and transport nutrients throughout their bodies.

FIGURE 4.1.5 (a) Sponges such as the azure vase sponge (*Callyspongia plicifera*) are (b) organised at the cellular level, with different types of cells performing different functions. Although the cells work together, they do not form true tissues or organs. Sponges are often referred to as tissueless multicellular organisms.

Despite the simple organisation of these organisms, each of the different cell types found within them has a specialised function that contributes to their survival and reproduction. These functions are outlined in Table 4.1.1. An example of a highly specialised cell in a simple multicellular organism can be seen in Figure 4.1.6 on page 104.

TABLE 4.1.1 Structure and function of specialised cell types in sponges

Cell type	Function	Structure
epidermal cells	protect the inner layer of cells	• thin, leathery • closely packed together
collar cells	• move water through the sponge's pores and into the central cavity (spongocoel), using the motion of their flagella • absorb nutrients	• flagella • hollow 'collar'
amoebocytes	• ingest and digest food caught by the collar cells • transport nutrients to the other cells of the sponge	mobile and flexible

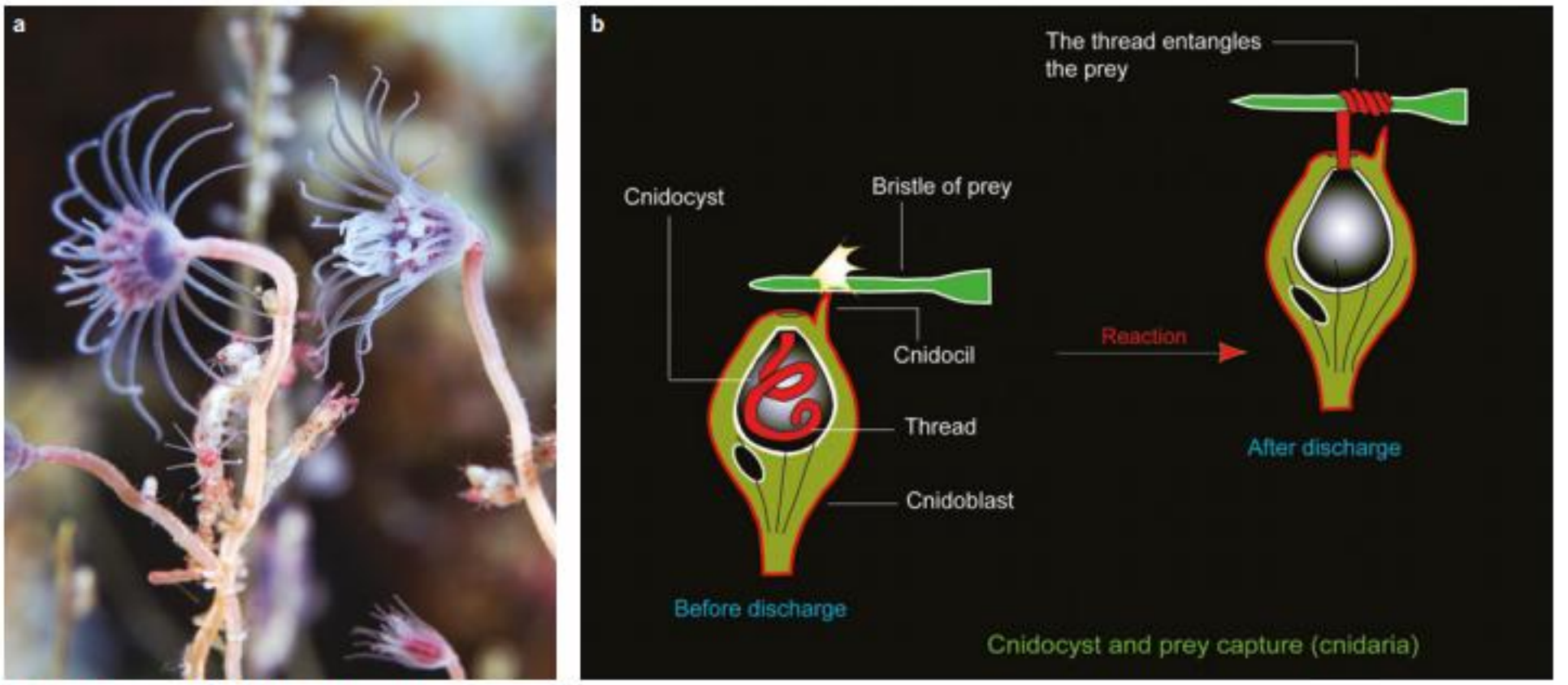


FIGURE 4.1.6 Simple multicellular organisms often have complex, specialised cells. (a) This cnidarian (*Ectopleura larynx*) is a marine animal with cells along its tentacles called cnidocytes or nematocytes. These cells are specialised to capture prey. (b) A thread is fired from the cnidocyst, a capsule within the cnidocyte, wrapping around and trapping prey. Other cnidarians, such as sea jellies and anemones, have cnidocysts that contain toxins that sting and paralyse prey.

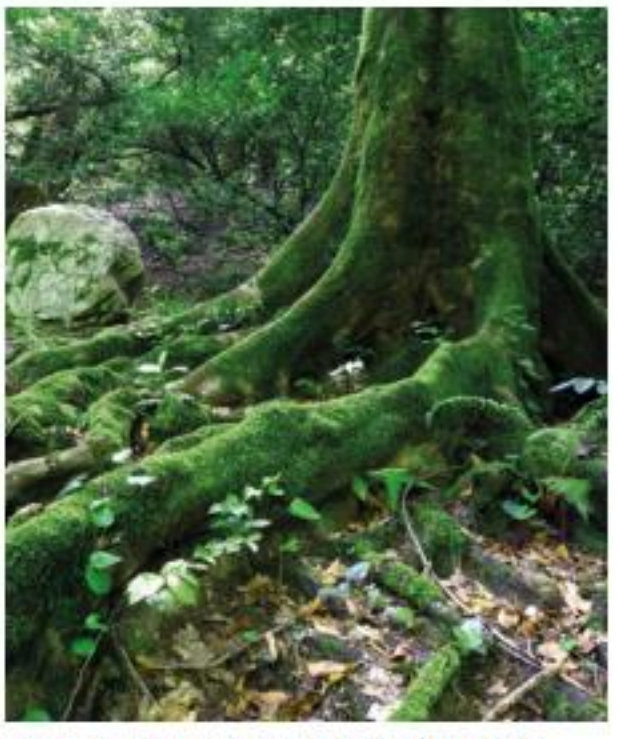


FIGURE 4.1.7 Moss is a non-vascular plant, whereas trees are vascular plants. Non-vascular plants lack vascular tissue and true organs, which reduces their ability to transport substances around the entire organism and limits the size to which they can grow. The vascular systems in vascular plants, however, enable trees to grow to great heights.

ORGANISATION IN COMPLEX VASCULAR PLANTS

A cellular level of organisation cannot meet the needs of larger and more complex organisms such as vascular plants. Consequently, cells in complex plants such as angiosperms (flowering plants) and conifers are organised into higher levels of organisation: tissues, organs and systems.

In comparison, non-vascular plants, such as algae and the moss in Figure 4.1.7, do not have vascular tissue or true organs. Instead, they have simplified tissues and absorb water directly through their cell walls, transporting it between cells by osmosis. The absence of vascular tissue in non-vascular plants limits their size owing to the lack of structural support and limited area over which they can transport water and nutrients.

Specialised cells in vascular plants

Specialised cells are cells that carry out a specific function. Some of the most important functions in vascular plants are involved in the transport of nutrients and water, and acquiring energy via photosynthesis. There are many specialised cells within the vascular tissue of plants for these functions. You will learn more about these specialised cells in Chapter 5.

Tissues in complex plants

The characteristic tissues in vascular plants (and the basis of this type of plant's name) are the vascular tissues, which are involved in the transport of water and nutrients throughout the plant. There are two types of vascular tissue: xylem and phloem.

Organs in complex plants

The major organs of vascular plants are as follows:

- Roots—responsible for absorbing and storing water and nutrients (mineral ions) required by the plant from the soil. Roots also function to support and anchor the plant to the ground. Root systems are often extremely complex and can be much larger than the above-ground structures of the plant. The large root systems of many trees in nutrient-poor rainforest soils do not penetrate deep into the soil layers and instead grow above ground (Figure 4.1.8).

- Leaves—the primary organ of photosynthesis. Photosynthesis is carried out to convert light energy into the chemical energy that fuels the organism's cells. The overall shape and organisation of leaves makes them well suited for their purpose. The major tissues of a leaf are the epidermis, photosynthetic tissue and vascular tissue. The vascular tissue (xylem and phloem) is visible as veins in the leaf structure (Figure 4.1.9).
- Stems—primary functions of the stems are to support the plants leaves, flowers and fruit; store nutrients; transport water and nutrients between the roots and the shoots; and grow new plant tissue. The stem is made up of three tissue types: dermal tissue, ground tissue and vascular tissue. The structure of stems varies widely between different species. The stems of strawberry runners are flexible and fleshy, while the stem or trunk of an oak tree is thick and woody. Some stems are even edible, such as asparagus and celery stalks.
- Flowers—the reproductive structures found in angiosperms. The function of a flower is to facilitate the fertilisation of the ovules (contained within the ovary) by the sperm (contained within pollen). The structures of many flowers are highly specialised to attract pollinators, such as bees, moths and fruit bats, to disperse the pollen from one flower to another. Other flowers produce pollen that is specialised for wind dispersal. After fertilisation, the seeds develop and the surrounding ovary grows into a fruit.
- Fruits—protect the developing seeds of the plant and help seeds to disperse from the parent plant. Fruits develop from the mature ovaries of flowers and often have a fleshy outer layer that surrounds the seeds. The outer structure of the fruit is often specialised to attract animals that aid in the dispersal of the seeds. Some animals, such as birds, eat the fruit and later excrete the seeds, while other animals disperse seeds that have attached to their fur. Examples of fruits are berries, peaches, tomatoes, nuts and legumes.

FIGURE 4.1.8 Two major organs of vascular plants are the leaves and roots, both of which are visible in this Amazonian tree with exposed buttress roots.

FIGURE 4.1.9 Several of the major organs of vascular plants can be seen on this orange tree, including leaves, flowers, fruits and stems. Note the visible veins in the leaf structure.

Systems in complex plants

Vascular plants have two systems: the root system and the shoot system. The root system is usually underground and functions to support the structure of the plant and absorb water and nutrients from the soil. The shoot system is made up of two parts: the non-reproductive (vegetative) parts of the plant, such as leaves and stems, and the reproductive parts, such as flowers and fruits. You will learn more about vascular plants in Chapter 5.

ORGANISATION IN COMPLEX ANIMALS

The animal kingdom includes the most complex types of multicellular organisms. An organ level of organisation is not enough to meet the needs of the most complex animals. For this reason, specialised cells of complex animals are organised into tissues, organs and systems.

Specialised cells in complex animals

Most complex animals are made up of hundreds of different cell types that are specialised to perform different functions. The roles of these cells are critical to the healthy functioning of the tissue, organ and organ systems of animals. As with vascular plants, there are many different types of specialised cells in complex multicellular organisms, and you will learn more about the specialised cells involved in obtaining nutrients, exchanging gases and removing wastes in Chapter 5.

Muscle cells

Muscle cells are specialised cells that can shorten or contract. Muscle cells can also lengthen or stretch in response to stimuli. In mammals, such as humans, there are three types of muscle cells: skeletal, cardiac, and smooth (Figure 4.1.10). While each of these muscle cells have similar properties, they have very different functions.

Skeletal muscle cells are part of the musculoskeletal system and allow for voluntary movement. Smooth muscle cells are found in internal organs, such as the intestines and arteries. Smooth muscles allow involuntary movement that is necessary for organs and systems to function. Cardiac muscle cells are specific to the heart. These cells are responsible for the contraction and relaxation of the heart.

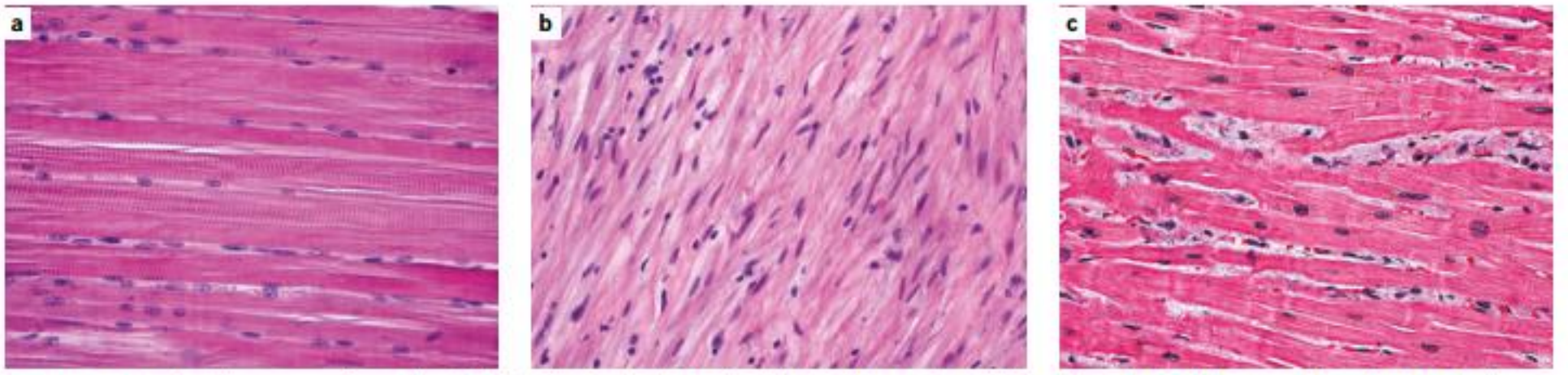


FIGURE 4.1.10 The three types of muscle cells in the human body are (a) skeletal, (b) smooth, and (c) cardiac. Each of these muscle cells share similar properties but have different functions in the body.

Tissues in complex animals

Specialised cells working together to carry out a specific function are called a tissue. For example, the function of a human red blood cell is to absorb and release oxygen as it travels around the body. However, one red blood cell cannot possibly carry all the oxygen that a human body needs. Billions of red blood cells need to work together to meet the needs of a human.

Cells do not need to be identical to be considered a tissue; they just need to be working together to carry out a particular function. For example, blood is a tissue that consists of red blood cells, white blood cells and platelets all working together.

Tissues in vertebrates are grouped into four basic types. These are shown in Figure 4.1.11.

- Muscle tissue is formed by cells that can contract (for example, skeletal and cardiac tissue).
- Nerve tissue consists of highly specialised cells called neurons that sense stimuli and transmit signals. This is essential for communication and coordination in complex multicellular animals. A motor neuron is shown in Figure 4.1.12.

- Connective tissue forms the supporting and connecting structures of the body (for example, bone and blood).
- Epithelial tissue consists of one or more layers of cells that cover most internal and external surfaces of the organism (for example, skin and intestinal lining).
- You will learn more about the tissues involved in obtaining nutrients, exchanging gases and removing wastes in Chapter 5.

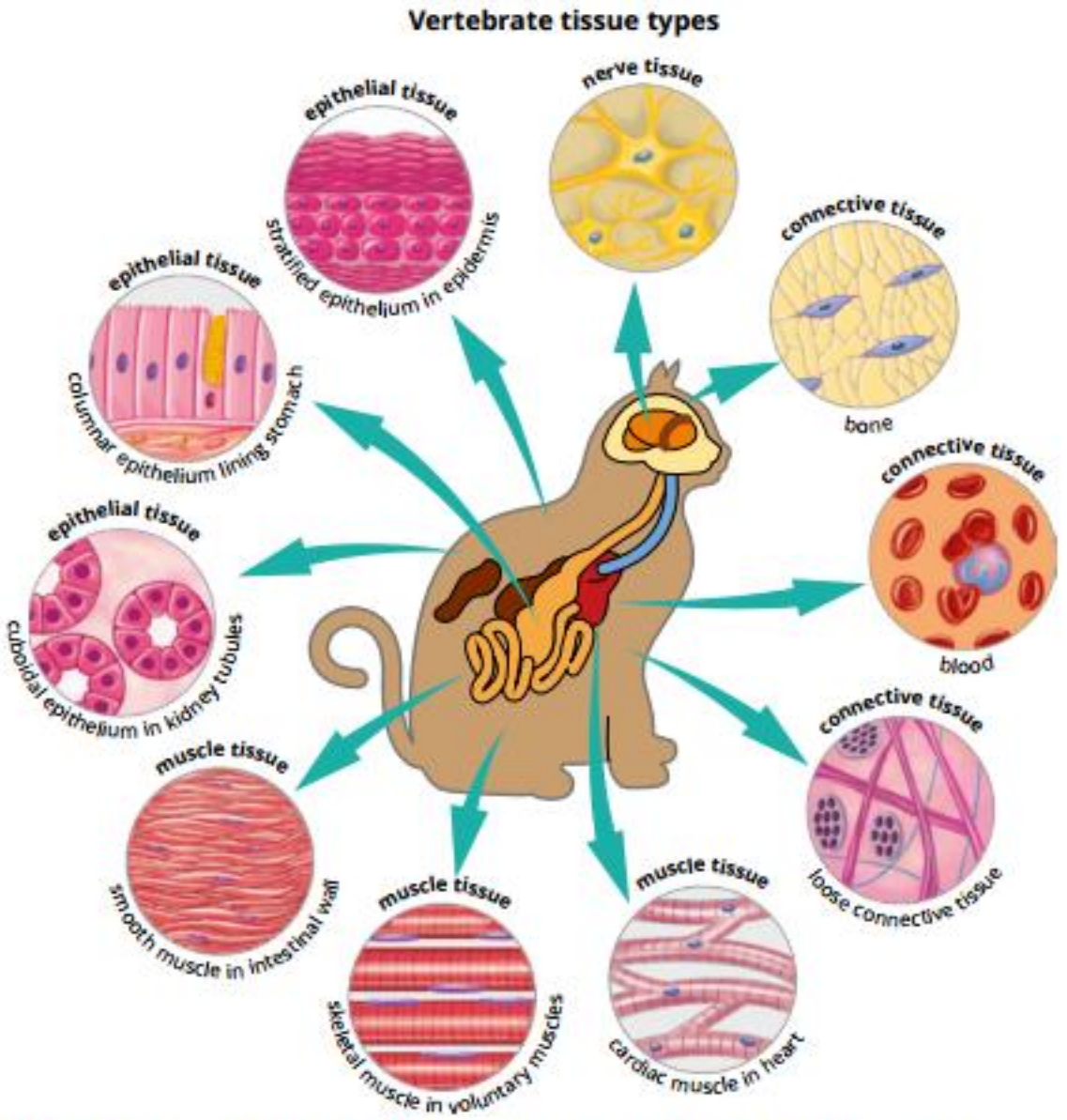


FIGURE 4.1.11 Complex multicellular organisms are made up of a diverse array of tissue types, specialised for many different functions. Vertebrate tissue types are shown here.

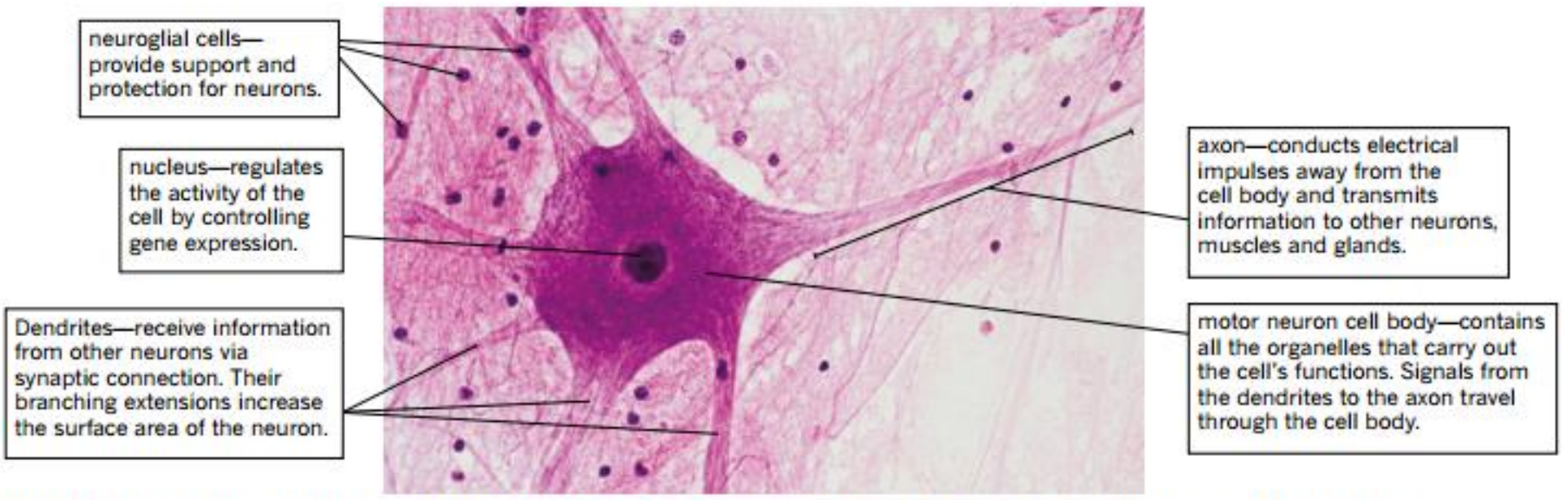

neuroglial cells—provide support and protection for neurons.
nucleus—regulates the activity of the cell by controlling gene expression.
Dendrites—receive information from other neurons via synaptic connection. Their branching extensions increase the surface area of the neuron.
axon—conducts electrical impulses away from the cell body and transmits information to other neurons, muscles and glands.
motor neuron cell body—contains all the organelles that carry out the cell's functions. Signals from the dendrites to the axon travel through the cell body.

FIGURE 4.1.12 A motor neuron cell and surrounding neuroglial cells from the spinal cord. These specialised structures are part of the tissue of the nervous system that is responsible for communicating signals that regulate and control bodily functions and activity in animals.

Organs in complex animals

An organ is a structure made up of two or more tissues that perform a specific function. Some of the many organs in complex animals include the eye, skin and heart. Below are descriptions of the eye and skin. You will learn more about the heart and other organs responsible for obtaining nutrients, exchanging gases and removing wastes in Chapter 5.

Eyes

The function of the eye organ is vision. Figure 4.1.13 shows an insect's eye. Insects have compound eyes that consist of thousands of individual units called ommatidia. Each ommatidium is like a single eye and collectively they are oriented to receive light from different directions, giving an insect a very wide angle of view. Each ommatidium consists of a lens, crystalline cone, light sensitive visual cells and pigment cells. The pigment cells ensure that light hits the visual cells at the correct angle. The visual cells transfer a message to the optic nerve, which transmits information to the brain.

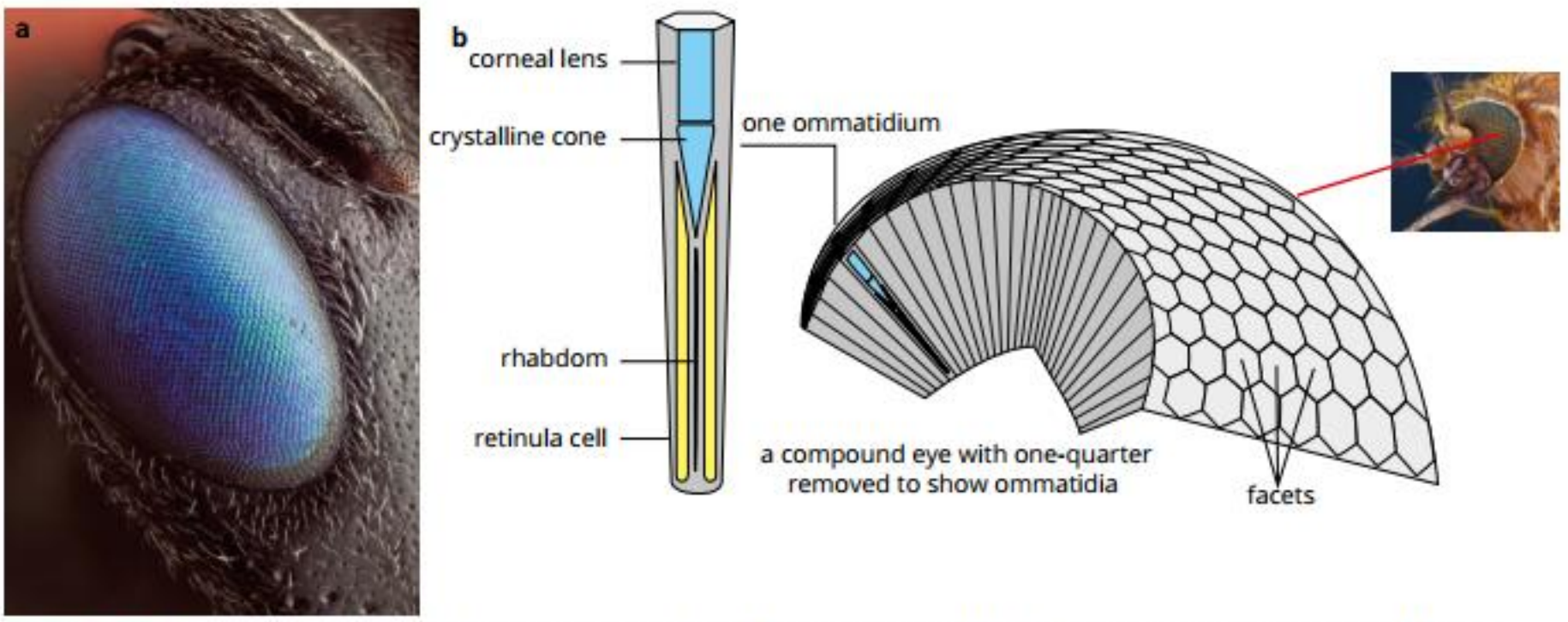


FIGURE 4.1.13 (a) A magnified view showing the external structure of the compound eye of the ensign wasp (*Evania appendigaster*). (b) The compound eye of a mosquito, showing the internal structure of the ommatidium typical of insects

Insect vision is different from human vision. The image generated is more like a light and dark mosaic, rather than a sharp image. Insect eyes are also capable of detecting very fast movement from a wide range of directions. This is why insects always seem to react very quickly to danger, such as a human hand.

Skin

The largest organ of the human body is the skin. In an average adult, the skin is more than 1.8 m^2 and makes up 6–10% of the body's total weight. The skin is considered to be an organ because it carries out a number of specific functions, including regulating temperature, preventing water loss and sensing the environment. Skin also provides a protective barrier and contributes to a stable internal environment for the other cells, tissues, organs and systems that make up a human.

Skin in humans is divided into three layers: epidermis, dermis and subcutis (the subcutaneous fatty layer). These layers are shown in Figure 4.1.14.

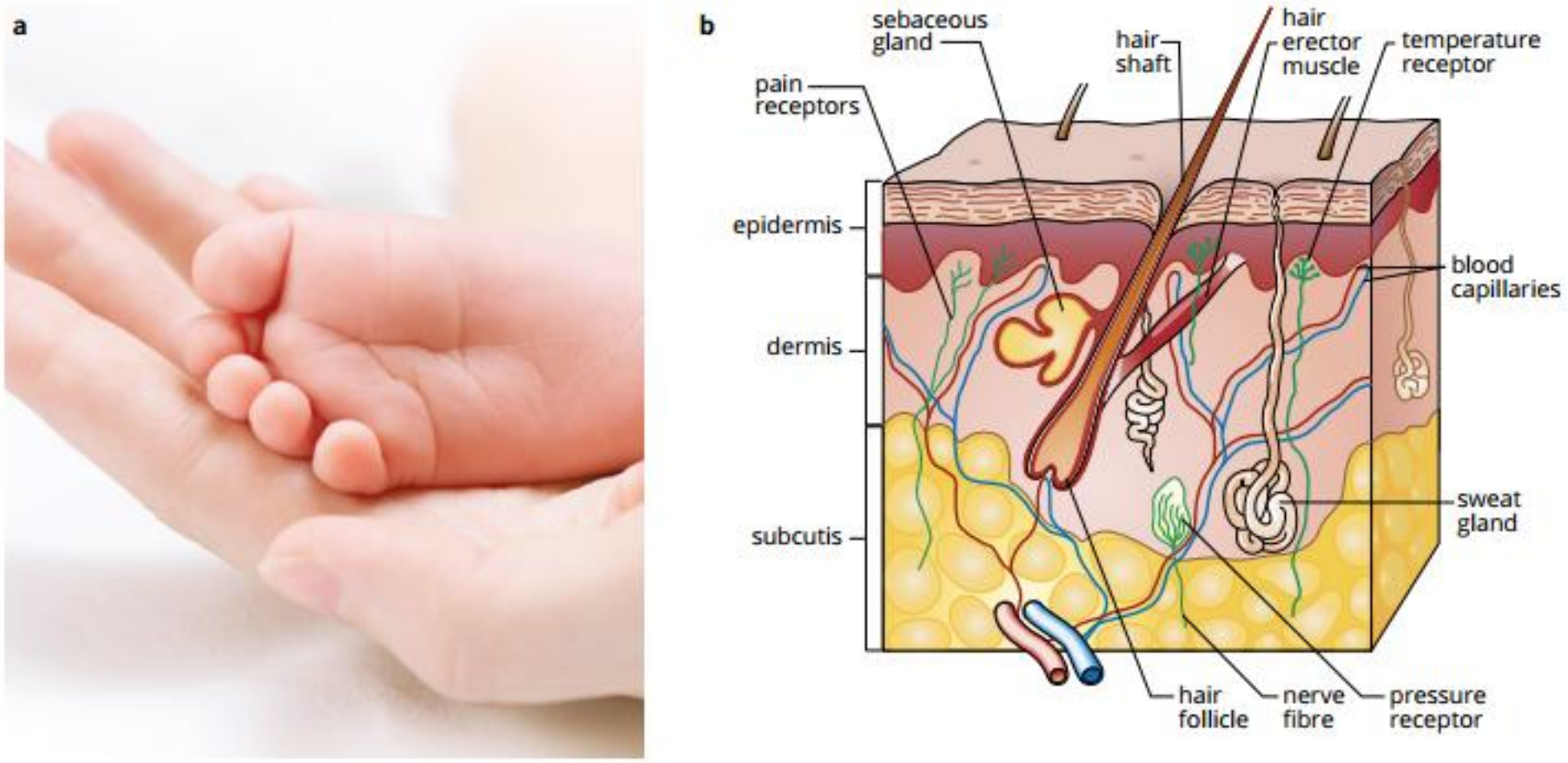


FIGURE 4.1.14 (a) Human skin is a complex organ. (b) Skin consists of many different types of specialised cells and tissues.

The epidermis is the outermost layer and consists mostly of keratinocytes. Keratinocytes are cells that contain keratin and they form a tough, waterproof layer for the body. They also prevent toxins, viruses and bacteria from entering the body. The outer layer of the epidermis consists of dead cells, which are continually replaced by dividing cells below. This layer of dead cells varies enormously over the body and is ten times thicker on the soles of the feet than on the face.

The dermis lays underneath the epidermis. It includes a range of nerves and receptors to sense external stimuli such as temperature, pressure and touch (see Chapter 6 for more details about receptors and stimuli). Some touch receptors are attached to hair cells, while pain receptors are close to the surface of the skin in the dermis or epidermis. Sweat glands aid in cooling by releasing a watery substance onto the epidermis via pores. Sebum is released by the sebaceous glands to help keep the skin and hair cells pliable. Sebum is also thought to have a mildly antiseptic effect on bacteria because of some of the fatty acid molecules present. The dermis has a rich supply of blood vessels that control blood flow through the skin, regulating the body temperature of the whole organism. The dermis also contains fibres of collagen and elastin, which give the skin strength and elasticity.

The deepest layer of human skin, the subcutis, consists mainly of fat cells. These fat cells act as a food reserve for the body and also provide insulation and cushion physical impact.

Systems in complex animals

Complex animals have multiple organ systems that work together to enable the animal to survive. Organ systems include:

- respiratory systems to exchange oxygen and carbon dioxide
- circulatory systems to transport substances throughout the animal's body
- reproductive systems to ensure continuation of the species
- nervous systems which control and coordinate the body and enable communication throughout the body.

You will learn more about organ systems in complex animals in Chapter 5.

4.1 Review

SUMMARY

- As an organism increases in size and complexity, greater cooperation and coordination is required between its cells.
- Multicellular organisms can be organised into the following levels to provide the needs of the entire organism:
  - specialised cells
  - tissues
  - organs
  - systems.
- Specialised cells are cells that have a specific function. Examples are neurons, red blood cells, skeletal muscle cells, smooth muscle cells, and cardiac muscle cells.
- Tissues are groups of similar cells working together to carry out a particular function in a multicellular organism.
- An organ consists of two or more tissues that work together to perform one or more specialised tasks. Examples are the heart and liver in mammals, and leaves and flowers in flowering plants.
- In multicellular organisms multiple organs work together to form an organ system. Examples in humans are the respiratory system and the circulatory system.

KEY QUESTIONS

Describe

1 List the levels of organisation in an organism in descending order, commencing with organism.

2 Describe the specialised function of muscle cells.

3 What is the primary function of the epidermis in multicellular organisms?

4 Recall the name of the two types of vascular tissue in plants.

5 Identify three examples of organs in vascular plants and describe each organ's function.

6 Define what is meant by the term 'specialised cell'?

7 Define 'tissue' (as used in biology) and provide an example of a tissue in a mammal.

8 Explain why the cells in multicellular organisms need to act in coordinated and organised ways.

Apply

9 Explain whether a finger is a tissue, an organ or an organ system.

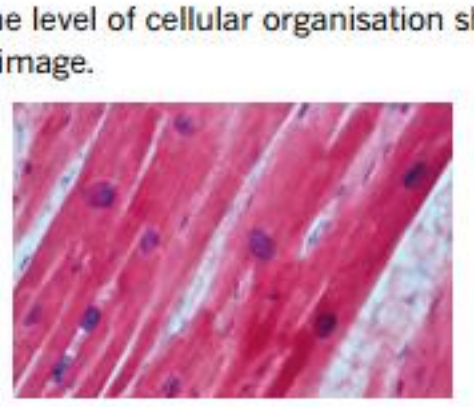
10 Infer why non-vascular plants tend to be smaller than vascular plants.

11 Infer why tissue-less multicellular organisms are often much smaller than more complex multicellular organisms.

12 A student argued that all animals had specialised cells. Explain whether you agree or disagree with this statement.

Analyse

13 Identify the level of cellular organisation shown in the following image.



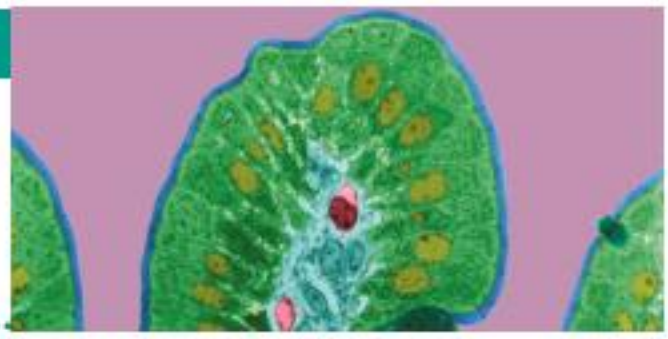
14 Identify the level of cellular organisation shown in the following image.

15 Identify the level of cellular organisation of the internal structures shown in the following image.

4.2 Stem cells and cell specialisation

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand how stem cells originate
- understand how cell specialisation occurs
- explain the different potential stem cells have to become new cells
- understand how and from where different stem cells are obtained.



Stem cells, like those in Figure 4.2.1, are cells that are yet to be specialised. In other words, they are unspecialised cells that have the potential to become different types of specialised cells. For this reason, scientists think that in the future stem cells, through innovative medical developments, could replace damaged cells that the human body cannot repair on its own, such as nerve cells. However, stem cell research is often associated with ethical concerns about the use of stem cells from **embryos**.

In this module, you will learn about how stem cells differentiate to become specialised cells. You will also discover the role and function of different types of stem cells.

FIGURE 4.2.1 A scanning electron micrograph of a clump of stem cells. This particular type of stem cell can differentiate into any of the cell types in the human body.

CELL SPECIALISATION

All multicellular organisms begin life as a single cell that resulted from the fusion of two highly specialised cells called gametes. Animal gametes are called the egg (or ovum) and sperm. The egg and sperm are shown in Figure 4.2.2. Plant gametes are called the ovule and pollen. Gametes are unique because they can fuse together to form a single cell, called a **zygote**.

This one cell, the zygote, contains all the genetic information required to develop into a fully functional multicellular organism. It is through cell replication, also known as **mitosis**, and cell differentiation that one single cell can become the trillions of highly specialised cells that make up an organism. Figure 4.2.3 shows a zygote replicating by mitosis.

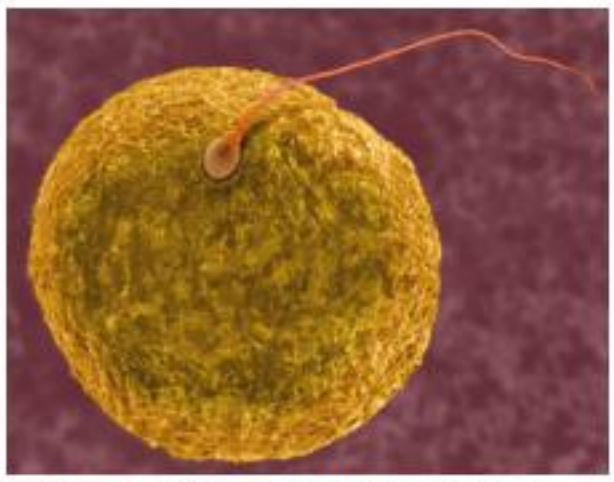


FIGURE 4.2.2 The egg (yellow) and sperm (orange) are two specialised cells. When fertilisation occurs, the egg and sperm form a single cell, called a zygote.

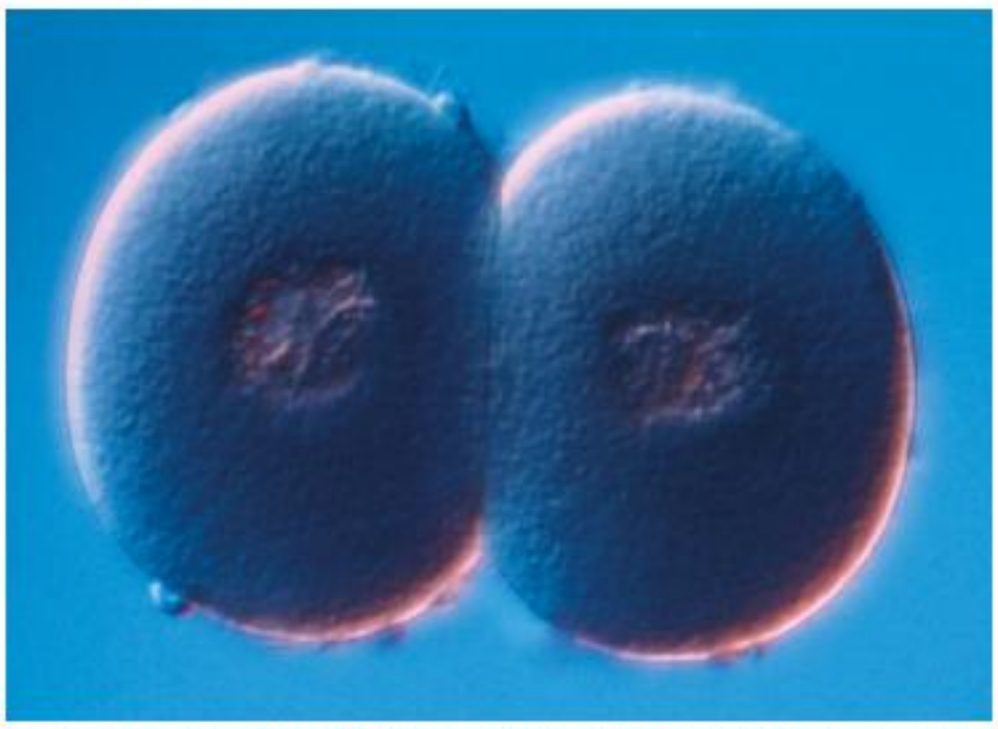


FIGURE 4.2.3 A zygote undergoing cell division, mitosis, which leads to the development of a multicellular embryo. At this stage, the zygote has undergone one cell division to create two cells.

Mitosis

Mitosis is one part of the cell cycle. It is a process where a cell replicates its chromosomes into two new nuclei which are then separated into two identical cells. Before a cell undergoes mitosis, it is in interphase, where each chromosome is replicated in preparation for cell division. Mitosis is a continuous process, but scientists have identified four key stages: prophase, metaphase, anaphase and telophase (Figure 4.2.4). After telophase comes cytokinesis. Cytokinesis is the physical process that splits the cell into two identical daughter cells.

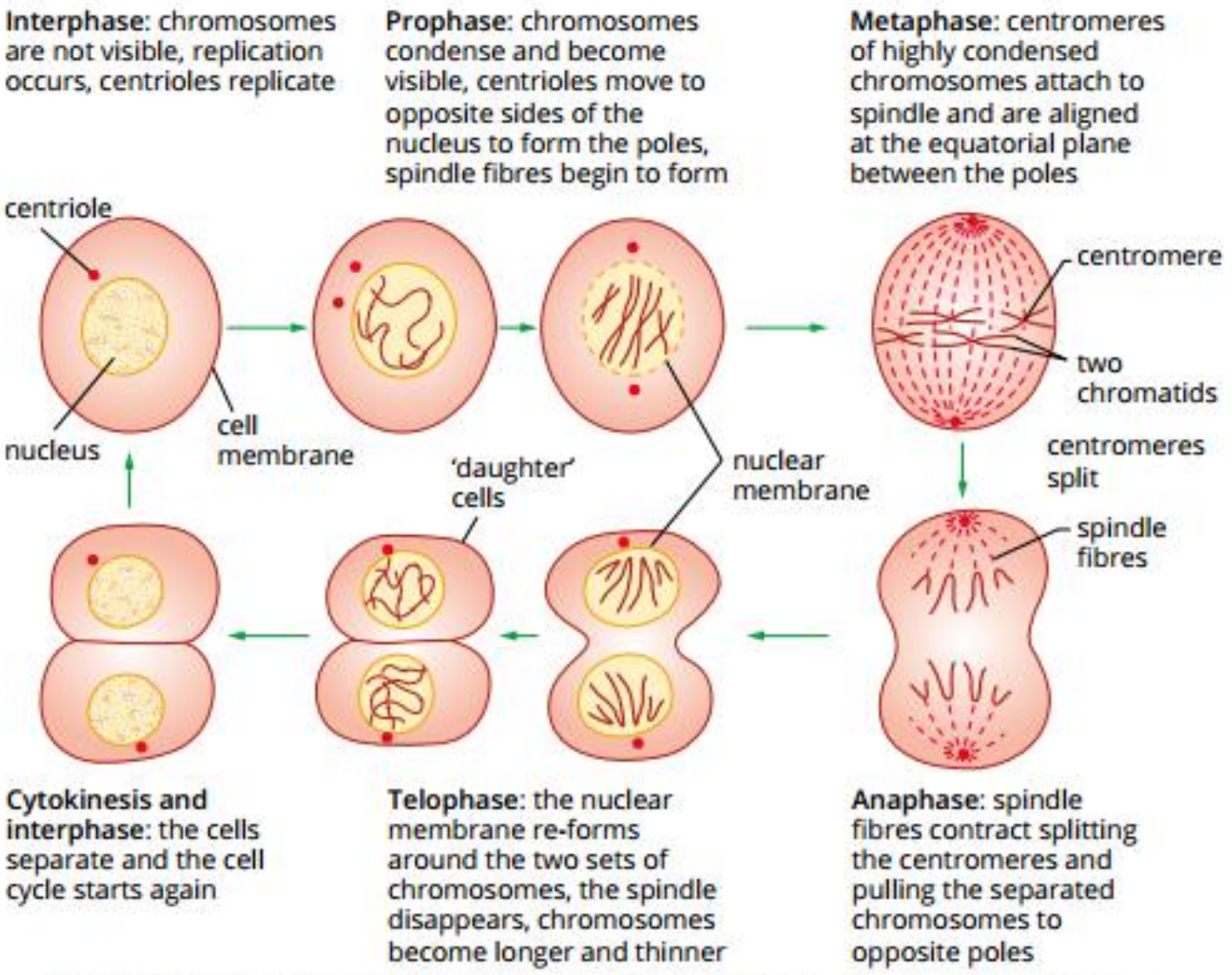

Interphase: chromosomes are not visible, replication occurs, centrioles replicate
Prophase: chromosomes condense and become visible, centrioles move to opposite sides of the nucleus to form the poles, spindle fibres begin to form
Metaphase: centromeres of highly condensed chromosomes attach to spindle and are aligned at the equatorial plane between the poles
centriole
nucleus
cell membrane
centromere
two chromatids
centromeres split
spindle fibres
nuclear membrane
'daughter' cells
Cytokinesis and interphase: the cells separate and the cell cycle starts again
Telophase: the nuclear membrane re-forms around the two sets of chromosomes, the spindle disappears, chromosomes become longer and thinner
Anaphase: spindle fibres contract splitting the centromeres and pulling the separated chromosomes to opposite poles

FIGURE 4.2.4 The four key stages of mitosis: prophase, metaphase, anaphase and telophase. Interphase is the stage of the cell cycle before mitosis.

Cell differentiation

Cell differentiation is the process by which unspecialised cells, **stem cells**, become specialised cells. It mostly takes place in all multicellular organisms. Stem cells are present in the embryo and some adult tissues of animals, and in meristem tissue in plants. Stem cells retain the ability to divide indefinitely by mitosis, while in specialised cells division is slowed or arrested, resuming to repair an injury or replace cells that have died.

Gene expression

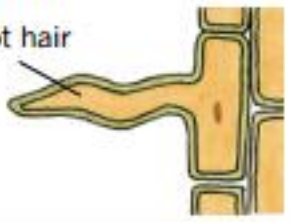
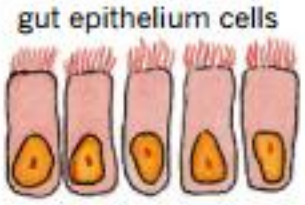
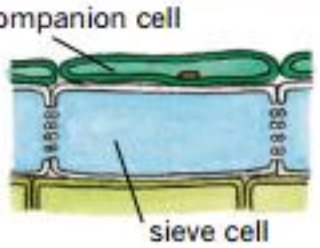
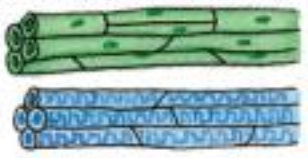
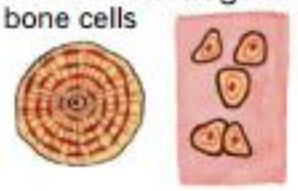
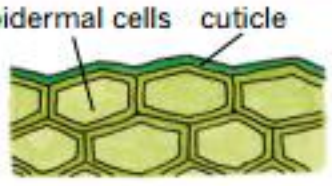
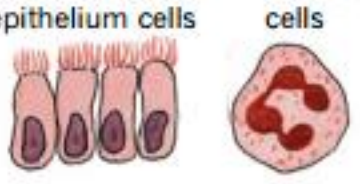
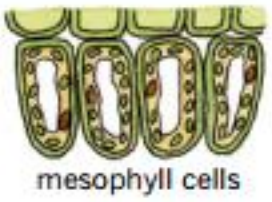
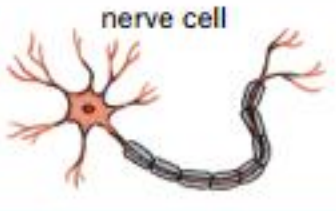
A stem cell will replicate via mitosis, but the resultant cells become specialised through gene expression. All of the genes required to produce every type of cell needed by an organism are present from fertilisation. Gene expression determines how a cell differentiates and functions.

Gene expression is the process in which the information stored in genes is used to build the different structures in a cell. In specialised cells, only some genes are active or expressed, and different specialised cells have different combinations of genes active and expressed. For example, in developing red blood cells, the genes for haemoglobin are expressed, while in gland cells the genes that code for different hormones, such as insulin, are expressed.

The internal and external structure of a cell is the basis for the functions it performs. Examples of the structures and functions of specialised cells in plants and animals are shown in Table 4.2.1. Cell specialisation is an advantage because cells are more efficient when they have only one function rather than many.

A nerve cell is specialised to carry electrical signals rapidly over long distances. It could not do this if it also had to break down food to obtain nutrients or protect against disease. A red blood cell, which is essentially a bag of haemoglobin that carries oxygen around the body, also cannot protect the organism from invading bacteria, which is the role of white blood cells. In plants, different cells are specialised for photosynthesis, exchange of substances (with other cells or the environment) and fluid transport.

TABLE 4.2.1 The structure and function of some specialised cells in plants and animals

Cell function	Cell specialisation	
	plant cells	animal cells
exchange	root hair 	gut epithelium cells 
transport	companion cell  sieve cell	red blood cells
strength/support	fibres (xylem, phloem) 	cartilage cells bone cells 
protection/defence	epidermal cells cuticle 	ciliated epithelium cells white blood cells 
photosynthesis	 mesophyll cells	
movement		muscle cells
communication		nerve cell 

An example of animal cell specialisation: your skin

One of the highly specialised organs of the human body is the skin, which consists of highly specialised cells. The outer layer of skin (the epidermis) alone consists of four different cell types and these interact with many others. You can see a diagram of the skin in Figure 4.2.5. Nerve cells, red and white blood cells, muscle cells and gland cells all contribute to the correct functioning of the skin cells in their role of protection, thermoregulation and sensation.

The structures of the specialised skin cells can give you clues about their function. The keratin-producing keratinocytes are the most common type of skin cell, making up 90–95% of the epidermis. The flat, scale-like structure of keratinocytes (also called squamous cells) helps them in their role of maintaining the structural integrity of the skin and tight junctions between them form an effective barrier. Keratinocytes also interact with nerve cells, antigen-processing Langerhans cells, sensory-processing Merkel cells and melanin-producing melanocytes.

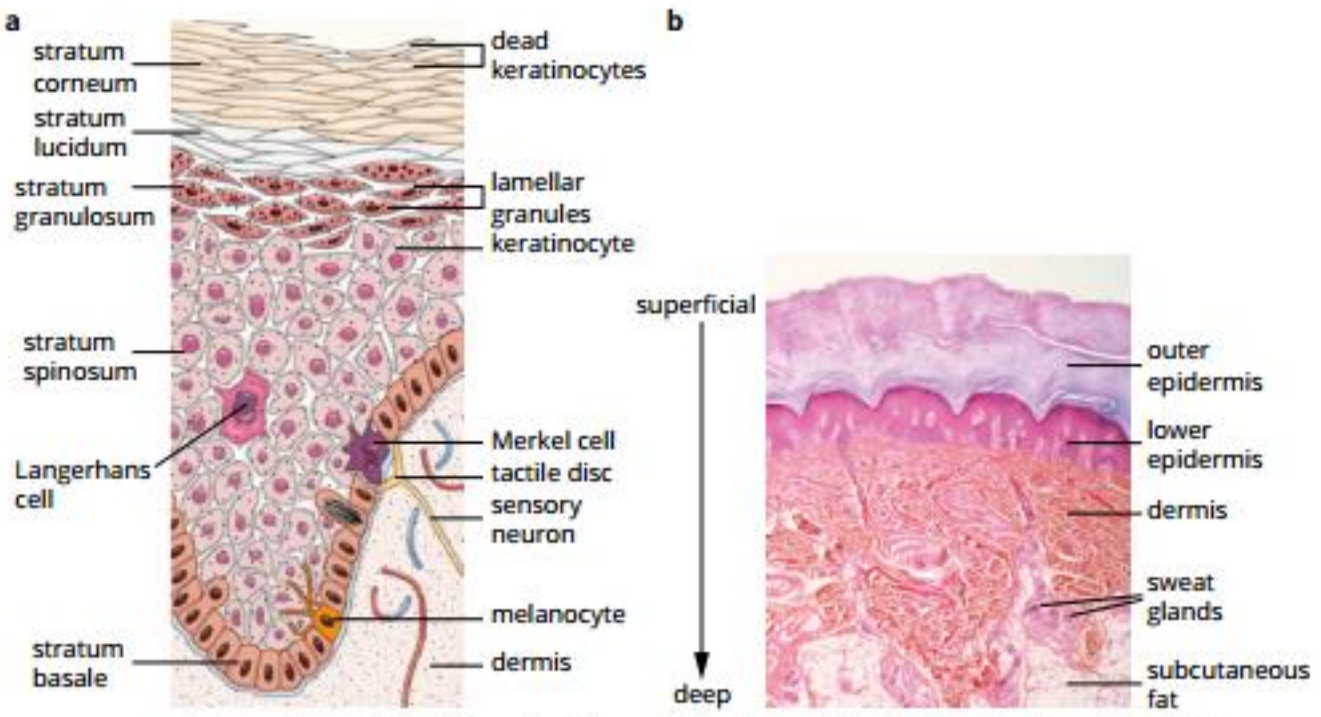


FIGURE 4.2.5 An overview of the different cell types that play a role in the functioning of the highly specialised epidermis, or upper layer, of human skin. (a) The epidermis consists of four principal cell types. (b) A photomicrograph of a portion of the skin

STEM CELLS

While all stem cells have the ability to differentiate into distinct cell types, the number of specialised cells a stem cell might differentiate into varies. Certain stem cells can divide indefinitely to replace cells, while others cannot.

Potency

Scientists characterise stem cells by their **cell potency**. Cell potency is a cell's ability to differentiate into other cell types. The more cell types into which a cell can differentiate, the greater its potency. Stem cells are classified as totipotent, pluripotent, multipotent or unipotent, depending on what sort of cells they can become:

- **totipotent** stem cells—are capable of giving rise to any cell type or a complete organism. A zygote and its divisions up to the 16-cell morula stage are the only human stem cells that are totipotent.
- **pluripotent** stem cells—can differentiate into any of the three germ layers: endoderm (e.g. lungs and gut lining), mesoderm (e.g. muscle, bone, blood), or ectoderm (e.g. skin and nervous system). Pluripotent stem cells cannot develop into a complete organism. These cells are present in the blastocysts. The primordial germ cells that give rise to gametes are also pluripotent.

The potency of a stem cell is indicated by the prefix, which comes from Latin.

toti = entire; total

pluri = many

multi = more than one

uni = one

- **multipotent** stem cells—have the ability to give rise to multiple, but limited, cell types. Haematopoietic (blood-forming) stem cells from the red bone marrow are an example of this cell type, as they can give rise to lymphocytes, macrophages, platelets and other blood cells.
- **unipotent** stem cells—can only differentiate into one cell type found in a specific tissue but can divide repeatedly. Skin epidermal stem cells are examples of unipotent cells that give rise only to new skin cells.

Stem cells may be also be characterised as either embryonic or adult. Embryonic stem cells are pluripotent and come from early-stage embryos. Adult stem cells are more differentiated than embryonic stem cells and so have less potency. Adult stem cells are multipotent or unipotent. The four potencies of stem cells and the life stages in which they are found are outlined in Figure 4.2.6.

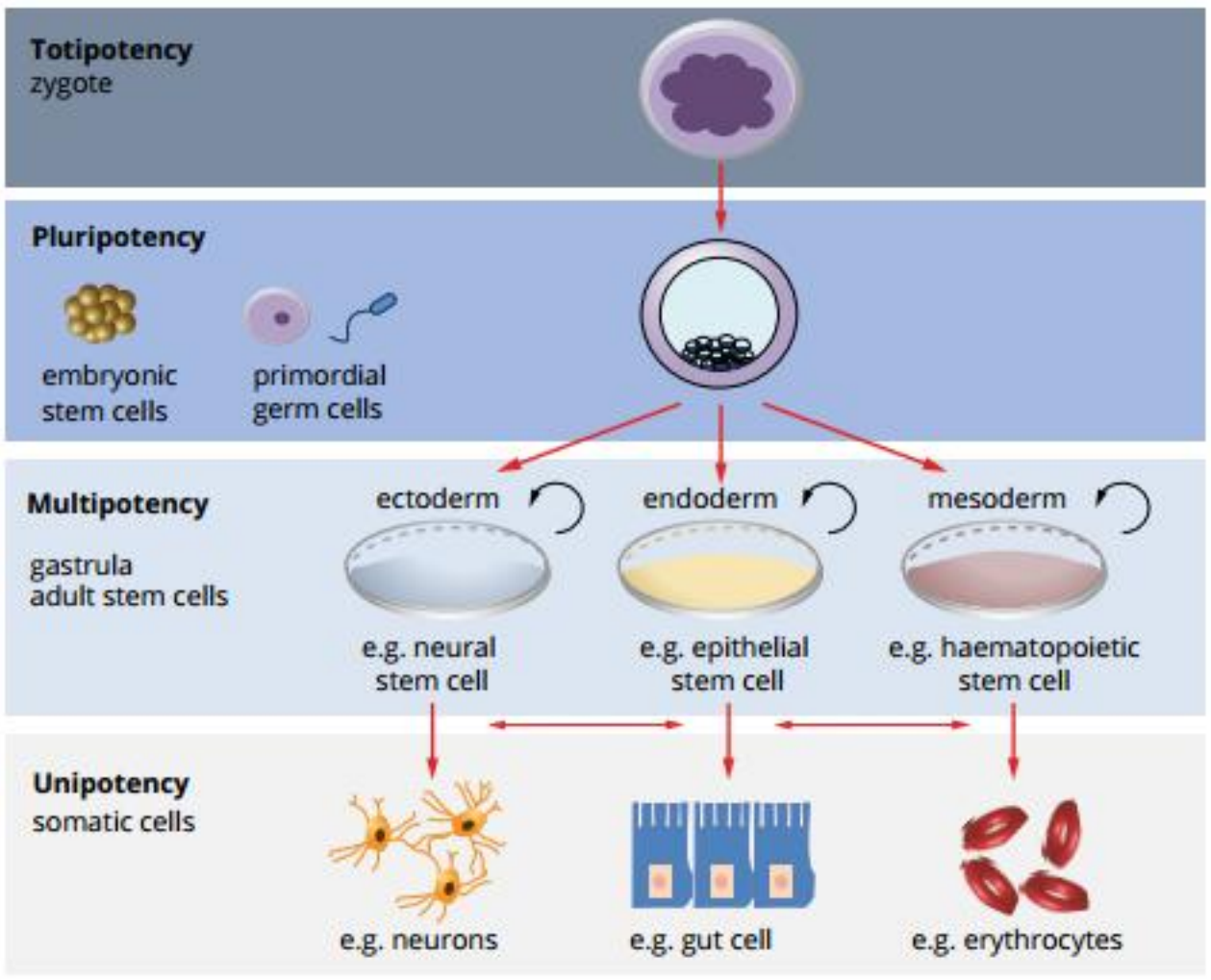


FIGURE 4.2.6 Stem cell potency is determined by how many cell types the stem cell can differentiate into. Totipotent cells have the greatest cell potency, followed by pluripotent cells, then multipotent cells and lastly unipotent with the least potency.

Embryonic stem cells

Embryonic stem cells are pluripotent and can replicate indefinitely. Embryonic stem cells are the undifferentiated or relatively undifferentiated cells of embryos (from the blastocyst stage). They can be obtained from surplus 3-day-old to 5-day-old embryos from in vitro fertilisation (IVF) programs. An understanding of embryo development is important to appreciate how and where the stem cells originated from.

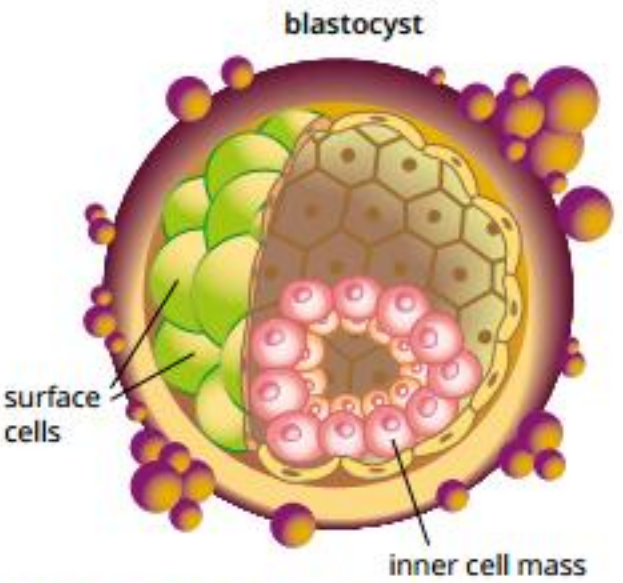


FIGURE 4.2.7 The blastocyst is characterised by an inner cell mass and an outer cell lining called the trophoblast.

Embryo development in humans

All humans start life as a single fertilised cell—a zygote. When the zygote undergoes mitosis to become two cells, it is no longer a zygote, but is now known as a **morula**. The morula continues to divide until it consists of 16 cells (after 3–4 days) and then enters the uterus. The morula is an early-stage embryo.

In the uterus, mitotic divisions continue, and the morula becomes a **blastocyst** (Figure 4.2.7) as cells begin to specialise. The blastocyst consists of a single layer of surface cells, which enable it to implant in the uterus and eventually develop into the placenta, and an inner cell mass that later gives rise to the embryo. After the blastocyst is implanted, **gastrulation** occurs over approximately 5 days, and the blastocyst becomes a **gastrula**, which has three different layers of cells—the ectoderm, endoderm and mesoderm. Eventually, the gastrula becomes an embryo, and then a **fetus** at three months (Figure 4.2.8).

The difference between each stage of embryo development is determined by the characteristics of the developing individual. As cells continue to specialise, structures and organs become more and more identifiable. For example, at about the eighth week of embryo development, the embryo is recognisable as a human, and is then known as a fetus, as shown in Figure 4.2.9. By the fetal stage, some of the internal and external organs and structures can be distinguished.

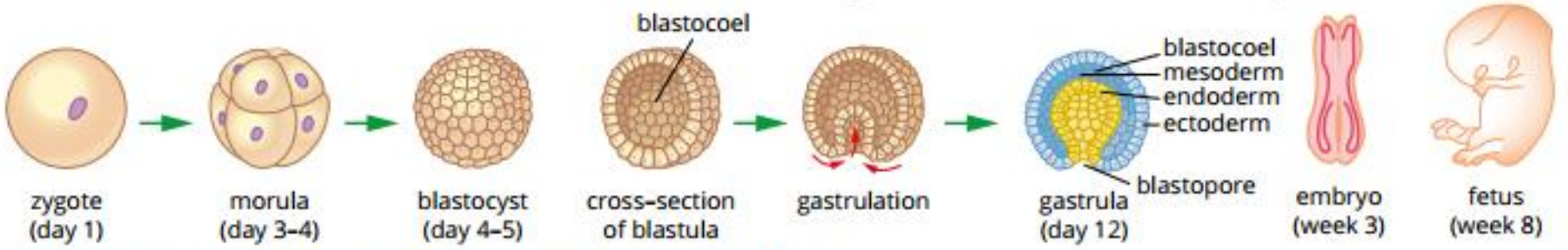


FIGURE 4.2.8 The development of a human zygote (fertilised egg) into a fetus

FIGURE 4.2.9 A human fetus at about the eighth week of development

- The brain, spinal cord and heart, which began development in the fifth week, continue to grow, the heart beats at about 150 beats per minute now and the lungs begin to form.
- Tissues grow that will become the spine and other bones.
- Eyes and ears begin to form.

Embryonic germ layers and cell specialisation

After implantation in the uterus, the blastocyst undergoes gastrulation, folding in on itself to form a gastrula with three primary layers of cells: ectoderm, mesoderm and endoderm. These primary layers are known as **germ layers** and are shown in Figure 4.2.10. The germ layers are also supported by two membranes:

- the yolk sac, which surrounds the egg yolk. It has a well-developed vascular system that transports nutrients from the egg yolk to the developing embryo
- the amnion, which surrounds the developing embryo, and is filled with fluid. Its main role is as a shock absorber to protect the embryo against any impacts or movements.

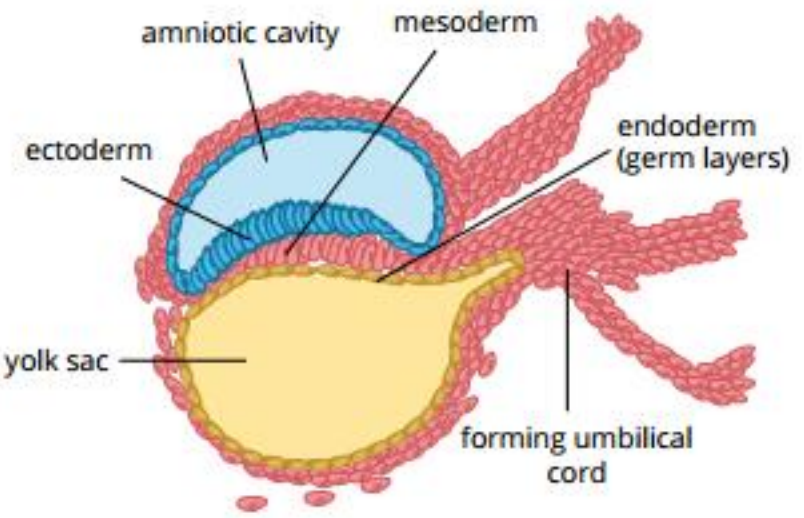


FIGURE 4.2.10 A gastrula, approximately 16 days after fertilisation, showing the three primary germ layers: ectoderm, mesoderm and endoderm

The three embryonic germ layers that form will eventually give rise to the different types of specialised cells that make up the tissues and organs in humans (Figure 4.2.11). These are the:

- **ectoderm** (outermost layer of the embryo), which forms epidermis, hair, peripheral nervous system, brain and spinal cord cells
- **mesoderm** (middle layer of the embryo), which forms muscle, cartilage, kidney and gonad cells
- **endoderm** (innermost layer of the embryo), which forms the lungs, bladder and lining of the digestive system, including the stomach, colon, liver and pancreas.

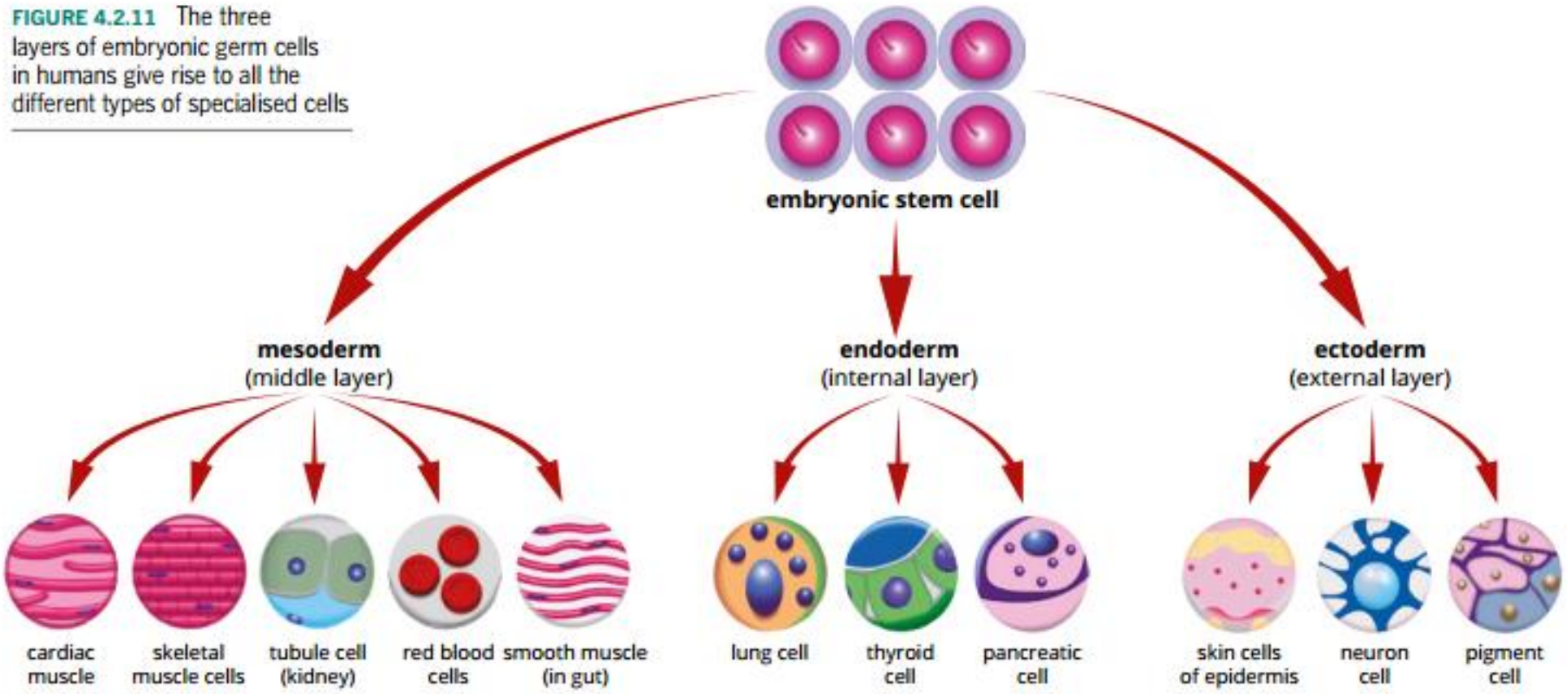


FIGURE 4.2.11 The three layers of embryonic germ cells in humans give rise to all the different types of specialised cells

Adult stem cells

Adult stem cells are present in small numbers in some adult tissues, such as hair follicles, bone marrow, the spinal cord and germ cells, and remain as stem cells throughout an individual's life. Adult stem cells can give rise to only a limited range of cells. For example, bone marrow (haemopoietic) stem cells, such as those in Figures 4.2.12 and 4.2.13 on page 118, can only produce blood cells. The biological purpose of adult stem cells is to repair and regenerate damaged and aged tissue, such as skin and liver cells. Adult stem cells cannot replicate indefinitely and are either multipotent or unipotent.

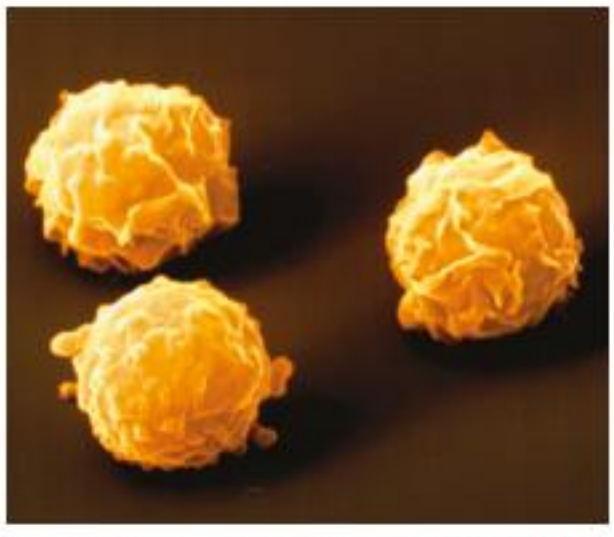


FIGURE 4.2.12 A scanning electron micrograph of human bone marrow stem cells. These adult stem cells can give rise to different types of blood cells only.

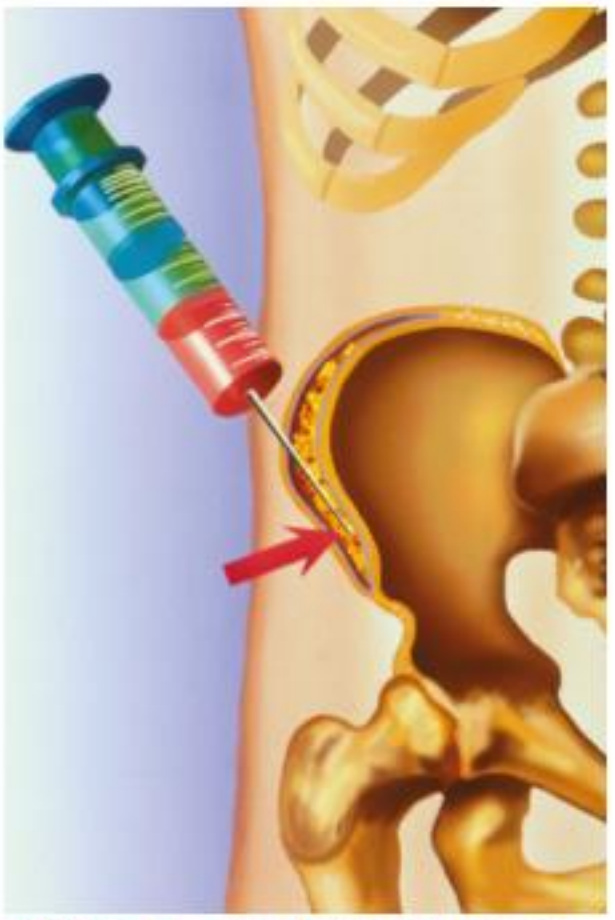


FIGURE 4.2.14 Harvesting bone marrow. The donor's bone marrow, which produces blood cells, is removed from the pelvis by syringe. The collected marrow is then filtered and treated before it is infused into the recipient.

FIGURE 4.2.15 A donated human placenta being prepared for the harvesting of umbilical cord stem cells.

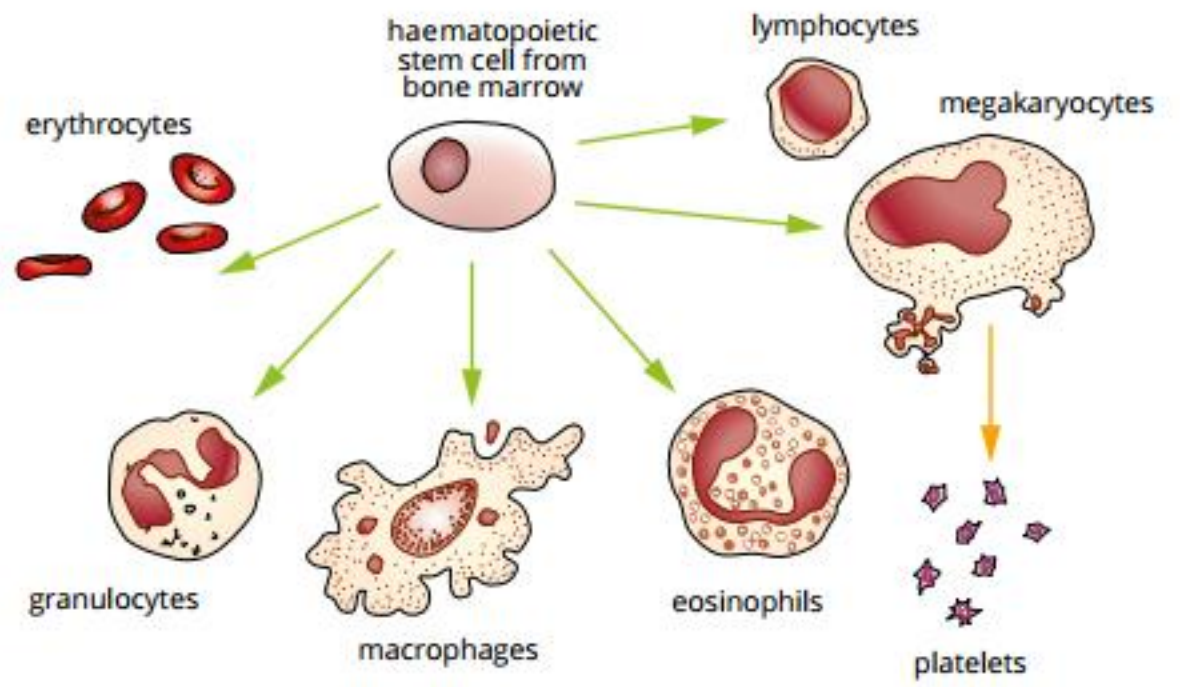


FIGURE 4.2.13 Haematopoietic (blood-forming) stem cells are set apart very early in development. They are found in bone marrow and are responsible for the continued production of a number of distinct cell types.

There are several sources of adult stem cells in the human body, including bone marrow and blood, cord blood, the intestines and the epidermis.

Bone marrow and blood stem cells

Bone marrow is the soft, fatty tissue found within bones. Bone marrow contains multipotent stem cells (called haematopoietic stem cells) that differentiate to give rise to all of your blood cells. Harvesting bone marrow or blood stem cells involves collecting the stem cells from a healthy patient. For bone marrow transplants, the bone marrow is taken from the pelvic region by inserting a large needle into the bone and withdrawing the marrow with a syringe. This procedure, which is shown in Figure 4.2.14, can be painful for the donor.

An easier and less painful procedure is to collect stem cells that are circulating in the blood. Peripheral blood stem cells are different from bone marrow stem cells, but the two stem cell types have similar degree of potency. To collect peripheral blood stem cells, a patient is connected to a machine and their blood is filtered to collect the stem cells, while the rest of the blood is returned to the patient.

Cord blood stem cells

Blood stem cells can also be taken from **cord blood**, which is blood taken from the umbilical cord after the birth of a baby (Figure 4.2.15). This blood contains 'adult' stem cells for the blood and immune systems in the body. The stem cells in cord blood are younger and healthier than stem cells from adult bones.

Intestinal stem cells

The lining of the intestines is a vital tissue in humans because it is the layer across which nutrients are absorbed into the body. The epithelial lining of the intestines has a remarkable ability to repair and renew itself because it contains adult stem cells. Scientists in the USA first harvested viable stem cells from the human intestine in 2013, and research into the possible uses of intestinal stem cells is continuing. Figure 4.2.16 shows a section of the small intestine with epithelial cells.

Epidermal stem cells

The skin is a very important organ because it regulates body temperature and acts as a barrier against pathogens. Like the intestines, it also has a remarkable ability to repair and renew itself after damage, using adult epidermal stem cells.

Skin stem cells are already being used to treat patients. Epidermal skin cells harvested from a person can be used to grow epidermis in a laboratory, as shown in Figure 4.2.17. These new layers of epidermis can then be grafted onto patients who have lost significant amounts of skin through, for example, third-degree burns.

FIGURE 4.2.16 A scanning electron micrograph of a section of the small intestine. The surface consists of deep folds, and the surface (epithelial) cells are shown in red. These cells are able to repair and renew very rapidly with the help of the adult stem cells present.

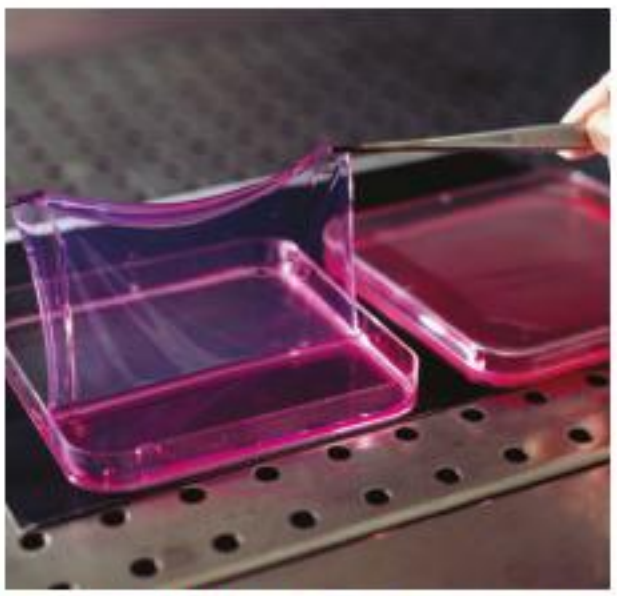


FIGURE 4.2.17 Artificially grown human skin being removed from a culture dish to make a skin graft. The cultures are made by transferring epithelial skin cells onto fibrin gel. The gel contains the nutrients needed by the cells to multiply and form skin. It takes only three weeks to grow a square metre of cultured skin.

Stem cells in other organisms

Stem cells are found in most adult organisms. Some organisms, such as the free-living planarians shown in Figure 4.2.18, retain a population of totipotent stem cells throughout their life, while other organisms, such as sea stars, retain a population of pluripotent stem cells throughout their life. These stem cells can develop into specific tissues, giving these organisms the remarkable ability to regenerate a body part that is completely lost through injury. This process is illustrated in Figure 4.2.18b and Figure 4.2.18c.

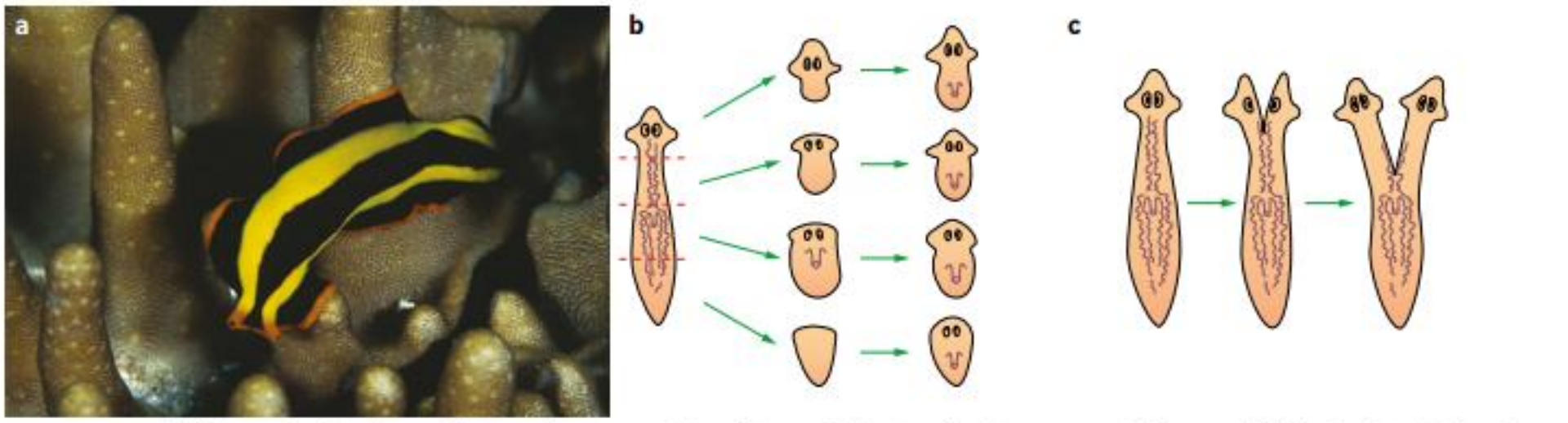


FIGURE 4.2.18 (a) This marine planarian, or flatworm, has a population of stem cells that enables it to regenerate if severed. (b) A planarian that has been cut transversely. (c) A planarian that has been cut longitudinally. Anterior regions are able to regenerate a new head faster than more posterior regions.

Unlike most animal cells, many cells in a plant's meristem tissue are able to continue differentiating and specialising throughout their entire life.

Plant cells have remarkable generative properties and display various levels of cellular potency. In plants, cell differentiation and cell specialisation derive from cells in the meristem tissue. Meristem cells are totipotent, and are located in the growing root tips, shoots, and buds of plants, as shown in Figure 4.2.19. Organs such as leaves and flowers develop from cells in the shoot apical meristem, while root growth comes from the cells of the root apical meristem. Scientific research also continues to examine how somatic cells can be reprogrammed to become pluripotent stem cells under specific conditions.

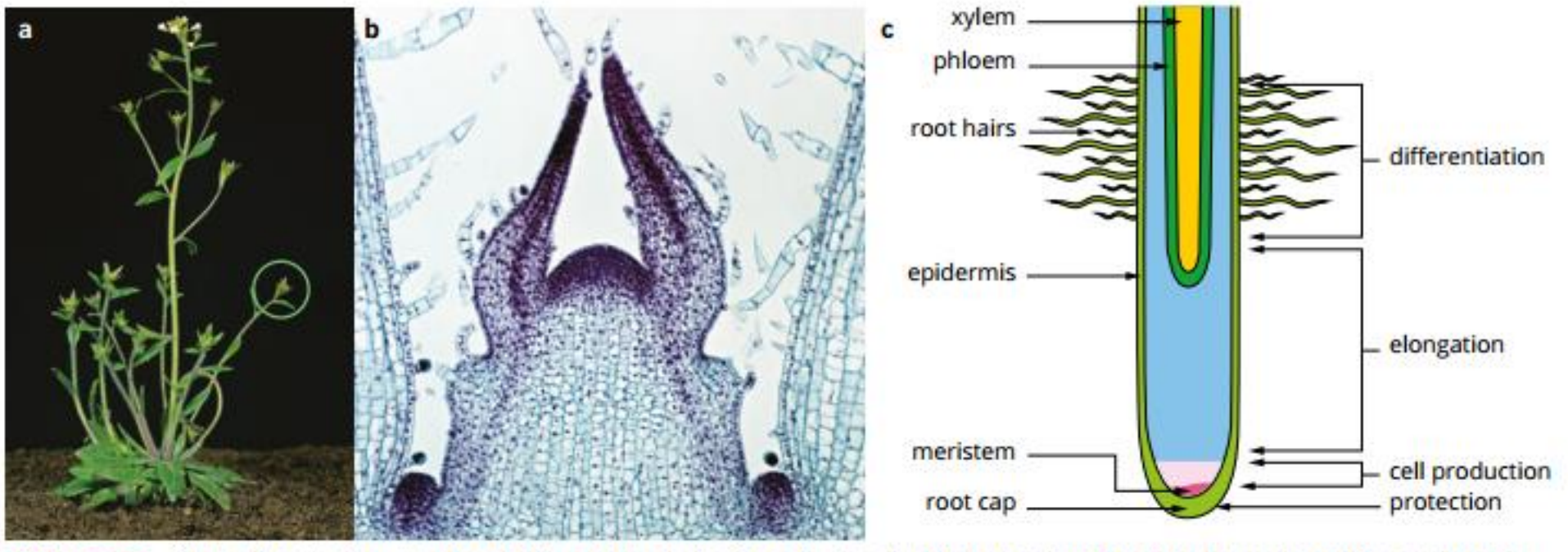


FIGURE 4.2.19 Plant cell production, growth and differentiation derives from developed, totipotent embryonic cells in the meristem. These cells are in the tips of shoots and roots in plants. (a) A thale cress (*Arabidopsis thaliana*) plant with the growing shoot tip highlighted. (b) Magnified image of stem cells in the shoot apical meristem. (c) The structures and regions of cell differentiation in the root meristem.

4.2 Review

SUMMARY

- All multicellular organisms start life as a single cell, known as a zygote.
- Cell replication (mitosis) and differentiation enable a single cell to produce all of the highly specialised cells in a multicellular organism.
- Cell differentiation is the process by which unspecialised cells, called stem cells, become specialised cells.
- Specialised cell function results from expression of particular sets of genes.
- There are two main types of stem cells: embryonic stem cells and adult stem cells.
- Embryonic stem cells can be thought of as 'all-purpose' cells that have the potential to develop into many different kinds of cells; stem cells are relatively undifferentiated cells.
- Stem cells can also be defined according to their potency, which refers to the number of different types of cells a stem cell can give rise to.
- Totipotent cells have the ability to become any cell type or an embryo. The human zygote is an example of a totipotent cell.
- Embryonic stem cells are pluripotent, which means they can differentiate into any of the three germ layers: endoderm (e.g. lungs and gut lining), mesoderm (e.g. muscle, bone, blood) or ectoderm (e.g. skin and nervous system). However, they cannot develop into a complete organism.
- Adult stem cells are classified as either multipotent or unipotent.
- Multipotent stem cells can give rise to multiple, but limited, cell types (e.g. haematopoietic stem cells can only give rise to blood cells).
- Unipotent stem cells can only give rise to one cell type in a specific tissue but can divide repeatedly.

KEY QUESTIONS

Describe

1 Recall the name of an undifferentiated cell that has the potential to become any cell type.

2 Recall where stem cells are present in:
- **a** plants
- **b** mammals.

3 Where in a plant can you find totipotent cells?

4 What is mitosis?

5 Explain the role of gene expression in cell differentiation.

6 Describe how embryonic stem cells differ from adult stem cells.

Apply

7 Classify the following cell types with their correct potency:
- **a** zygote
- **b** embryonic stem cell
- **c** adult stem cell

8 Explain the function adult stem cells play in repairing the lining of the human intestines.

Analyse

9 What is an advantage of harvesting peripheral blood stem cell over bone marrow stem cells?

10 Refer to Table 4.2.1 on page 113. Select a cell type and assess how its structure assists its function.

11 Axolotls, like the one in the image below, can regrow limbs if they get cut off. Identify the level of cellular potency of the stem cells found in axolotl limbs and explain what this means.

Chapter review

04

KEY TERMS

adult stem cell
blastocyst
cell cycle
cell differentiation
cell potency
cord blood
daughter cells
embryo
embryonic stem cell
epidermis
ectoderm
endoderm
fetus
gastrula
gastrulation
germ layer
mesoderm
mitosis
morula
multicellular
multipotent
organ
organism
pluripotent
specialised cell
stem cell
tissue
totipotent
unicellular
unipotent
zygote

KEY QUESTIONS

Describe

1 A fertilised egg is also known as:
- **A** a blastocyst
- **B** an embryo
- **C** a gastrula
- **D** a zygote.

2 Embryonic stem cells are:
- **A** totipotent
- **B** pluripotent
- **C** multipotent
- **D** unipotent.

3 The two systems in vascular plants are:
- **A** flower system and leaf system
- **B** fruit system and stem system
- **C** photosynthetic system and respiratory system
- **D** root system and shoot system

4 The stomach is an example of:
- **A** a specialised cell
- **B** a tissue
- **C** an organ
- **D** a system.

5 Explain the difference between a tissue and an organ.

6 Explain why large multicellular organisms require organ systems.

7 Explain the role of gene expression in cell specialisation.

Apply

8 Why is cell specialisation an advantage?

9 Label the three germ layers in the image of a gastrula below.

10 Planarians (flatworms) retain a population of totipotent stem cells throughout their life. Describe how this observation explains why a planarian, when cut in half, can regenerate.

11 Succulent plants can be easily propagated from cuttings. Explain this phenomenon in relation to stem cells.

12 Construct a flowchart that shows the levels of organisation in complex plants and animals, indicating how these groups of organisms are similar.

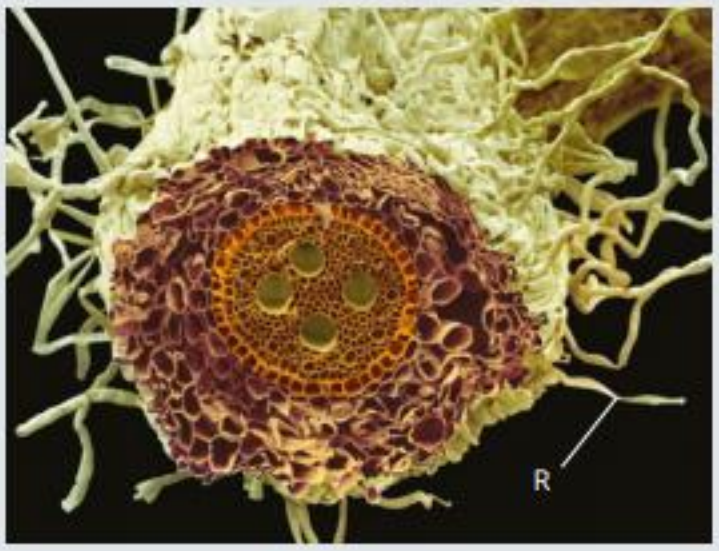
13 Using an example from the circulatory system, explain how the hierarchical organisation of cells, tissues and organs contributes to the survival and functioning of a human.

14 How do stem cells contribute to tissue repair and regeneration?

Analyse

15 Determine which level of cellular organisation best characterises the following image.

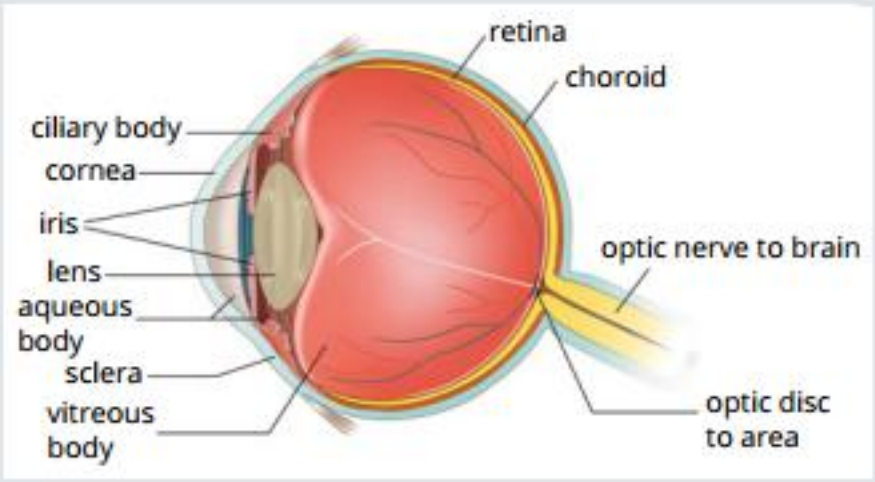
16 The following image shows a scanning electron micrograph of a root.



a Explain how cell R is specialised in its function.

b The root is an organ. Using the root as an example, outline how the cells are organised to form an organ.

17 The following image shows some of the parts of the human eye.



Corneal transplants enable the cornea to be replaced if it is diseased or damaged. Only the cornea can be transplanted, not the entire eye. Categorise the level of cellular organisation of the cornea.

18 A student argued that cell specialisation begins in human development during the zygote stage, as this is when some of the internal and external organs and structures can be distinguished. Explain whether you agree with the student.

19 Determine with reason why the order of embryonic development is from diagram **b** to **c** then **a**.

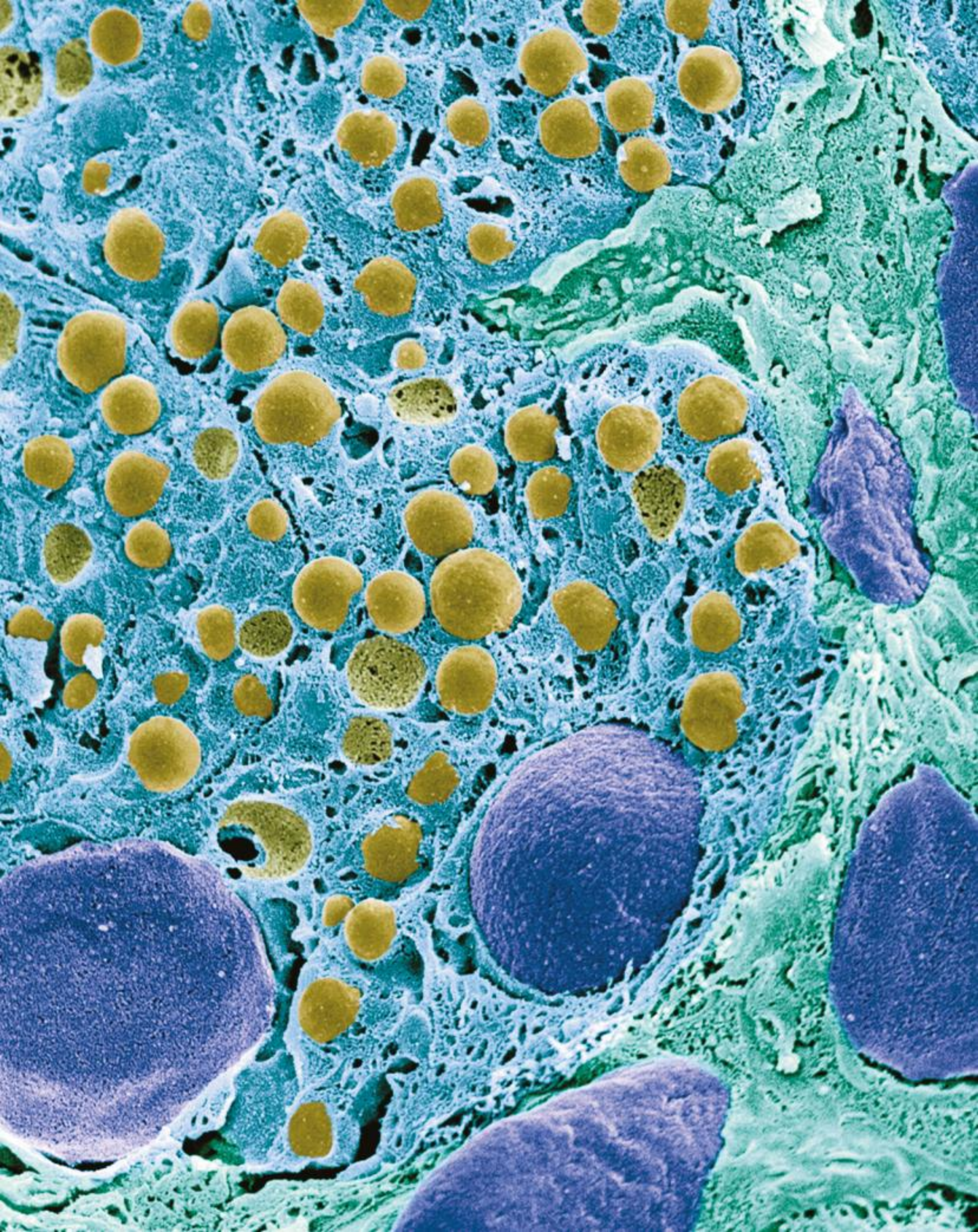
Interpret

20 Research a multicellular organism indigenous to your area and identify three specialised cell types. For each cell type:

- **a** explain how its structure relates to its function
- **b** list the tissue, organ and system the cell belongs to
- **c** identify the germ line from which the specialised cell originated.

21 Scientists are continuing to investigate ways to induce stem cells in animals. Review some recent research and summarise the findings, including:

- **a** which cells were the scientists experimenting on
- **b** what level of cellular potency they were able to achieve, if any
- **c** the new knowledge or benefits of the research.



CHAPTER

05 Organ systems

In this chapter, you will learn how multicellular organisms conduct the complex tasks of moving substances into and out of their bodies. The mechanisms via which products enter and leave an organism demonstrate not only the organism's level of complexity, but also the particular demands of the environmental niche in which the organism lives.

For all living organisms, gaseous exchange, ingestion and breakdown of nutrients, transport of essential products around the organism and removal of waste are all fundamental processes.

While the mechanisms that enable these processes vary between species, each process enables individual species to survive and compete successfully for resources in their environment.

While various systems are discussed separately, it is important to remember that many of the systems' functions overlap and that they work in a coordinated way for the survival of the organism as a whole.

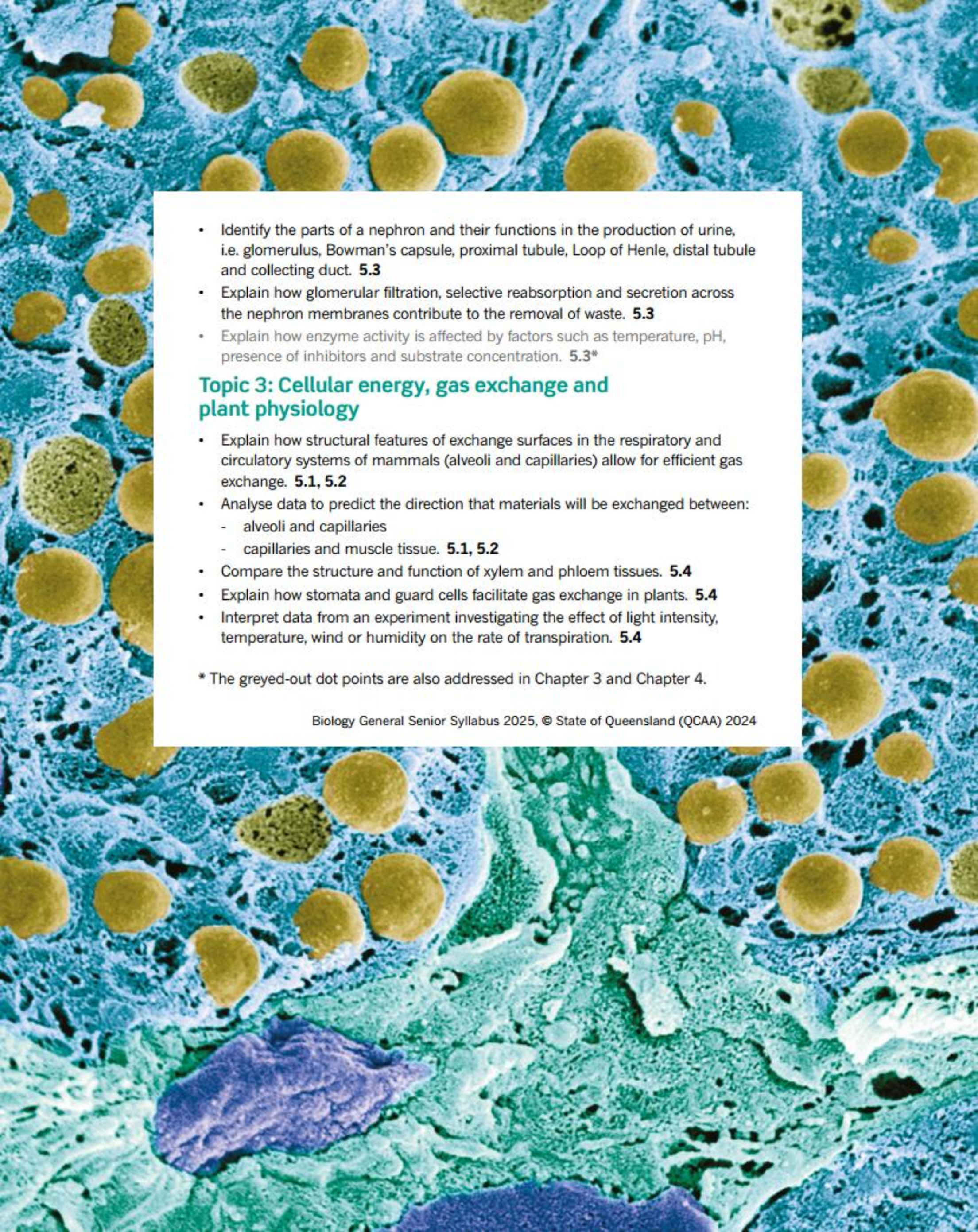
Syllabus subject matter

Topic 1: Cells as the basis of life

- Describe how the hierarchical organisation of cells, tissues, organs and systems allow multicellular organisms to
  - obtain nutrients, e.g. digestive and circulatory systems
  - exchange gases, e.g. respiratory and circulatory systems
  - remove wastes, e.g. respiratory, circulatory and excretory systems. **5.1, 5.2, 5.3***
- Explain that each body system contains specialised cells and tissues that are structurally suited to function, including:
  - size and shape (SA:V ratio)
  - organelle composition. **5.1, 5.2***
- Interpret data from an experiment investigating the effect of surface area-to-volume ratio on the rate of diffusion. **5.1, 5.2, Practical investigation 1**

Topic 2: Exchange of nutrients and wastes

- Describe the structure and function of carbohydrates, proteins and lipids. **5.3***
- Describe the role of amylase, protease and lipase in chemical digestion. **5.3**
- Explain how structural features of exchange surfaces in the digestive and circulatory systems of mammals (e.g. villi and capillaries) allow for efficient nutrient exchange. **5.3**
- Describe how closed circulatory systems facilitate the efficient transport of materials to and from all cells in the body. **5.1, 5.2**



- Identify the parts of a nephron and their functions in the production of urine, i.e. glomerulus, Bowman's capsule, proximal tubule, Loop of Henle, distal tubule and collecting duct. **5.3**
- Explain how glomerular filtration, selective reabsorption and secretion across the nephron membranes contribute to the removal of waste. **5.3**
- Explain how enzyme activity is affected by factors such as temperature, pH, presence of inhibitors and substrate concentration. **5.3***

Topic 3: Cellular energy, gas exchange and plant physiology

- Explain how structural features of exchange surfaces in the respiratory and circulatory systems of mammals (alveoli and capillaries) allow for efficient gas exchange. **5.1, 5.2**
- Analyse data to predict the direction that materials will be exchanged between:
  - alveoli and capillaries
  - capillaries and muscle tissue. **5.1, 5.2**
- Compare the structure and function of xylem and phloem tissues. **5.4**
- Explain how stomata and guard cells facilitate gas exchange in plants. **5.4**
- Interpret data from an experiment investigating the effect of light intensity, temperature, wind or humidity on the rate of transpiration. **5.4**

* The greyed-out dot points are also addressed in Chapter 3 and Chapter 4.

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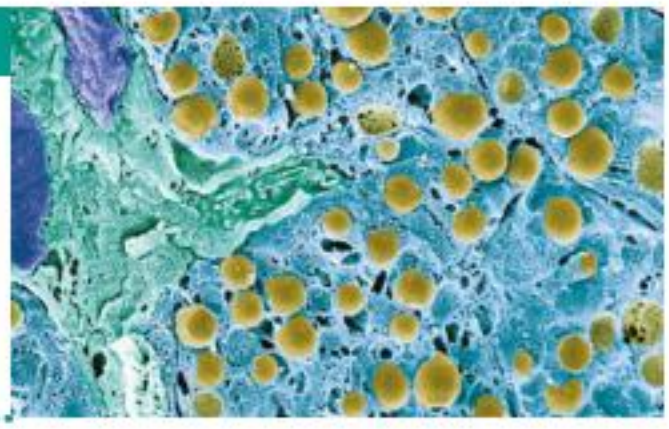
5.1 Animal transport systems

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain how the structure and function of capillaries enables efficient exchange of substances
- describe how open and closed circulatory systems function
- compare and contrast open and closed circulatory systems
- analyse the importance of the lymphatic system to the operation of the circulatory system.



The function of transport systems is similar in all organisms: to transport nutrients to cells and remove wastes from them. In simple multicellular organisms, nutrients are transported by diffusion between cells. However, diffusion only works over short distances from cell to cell. In large, complex animals, diffusion is not sufficient to deliver essential nutrients to all of the organism's cells. Moreover, not every cell in a multicellular organism has equal access to the external environment. It is for these reasons that specialised circulatory systems, with networks of pipes and chambers, have evolved to transport vital nutrients to all cells in complex multicellular organisms.

OPEN AND CLOSED CIRCULATORY SYSTEMS

There are two broad types of transport systems for multicellular organisms: **open circulatory systems** and **closed circulatory systems**.

Arthropods, including insects, have an open circulatory system. The primary function of open circulatory systems is to provide nutrients and remove wastes.

An open circulatory system has a heart or heart-like structure but no blood vessels (Figure 5.1.1). The blood-like fluid in an open circulatory system is called **haemolymph**. Haemolymph mostly consists of water and usually carries biomolecules such as proteins, lipids and carbohydrates, as well as wastes such as nitrogenous wastes.

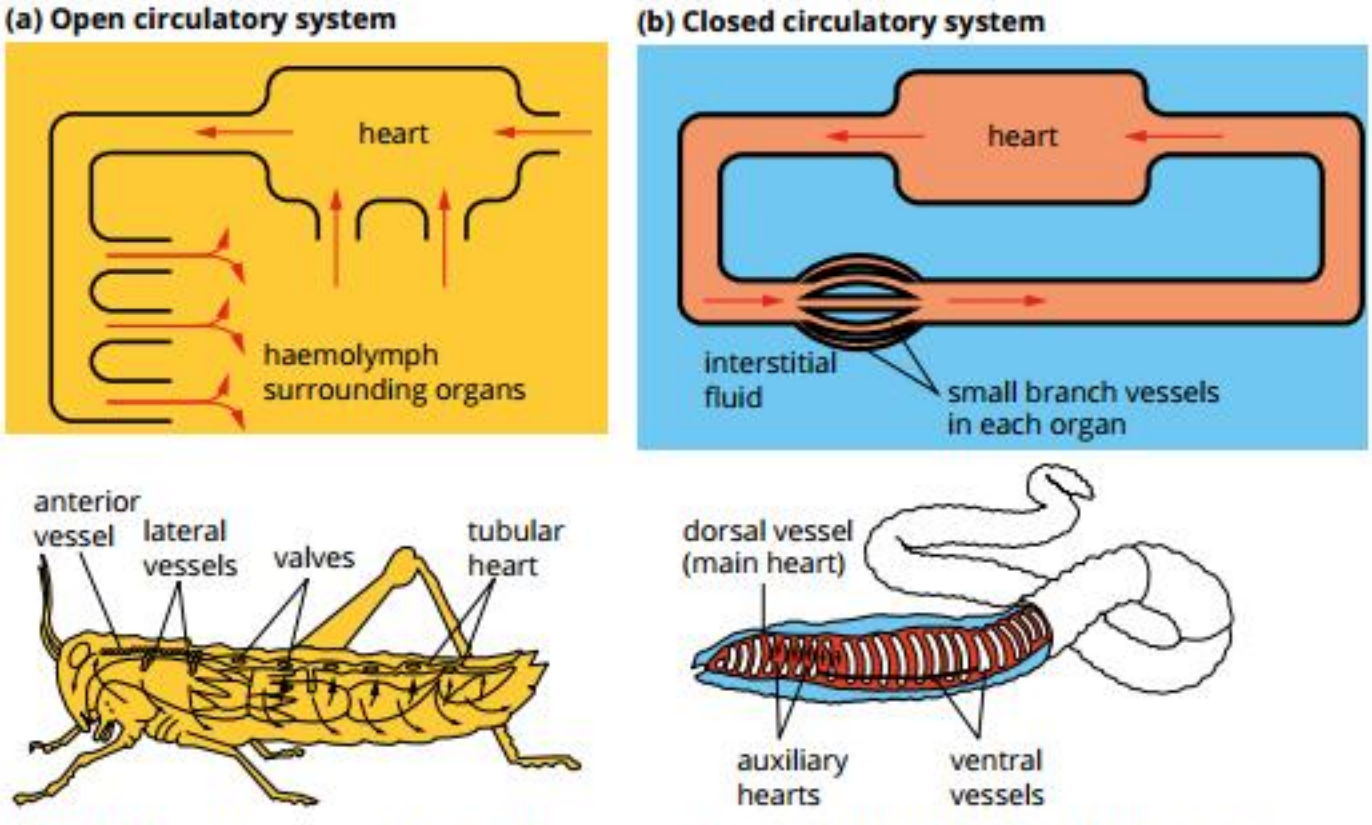


FIGURE 5.1.1 (a) The open circulatory system of a grasshopper and (b) the closed circulatory system of an earthworm have different structures but both systems have the same function. They supply the cells of the body with nutrients and oxygen and transport wastes away from the cells to the organs that excrete them.

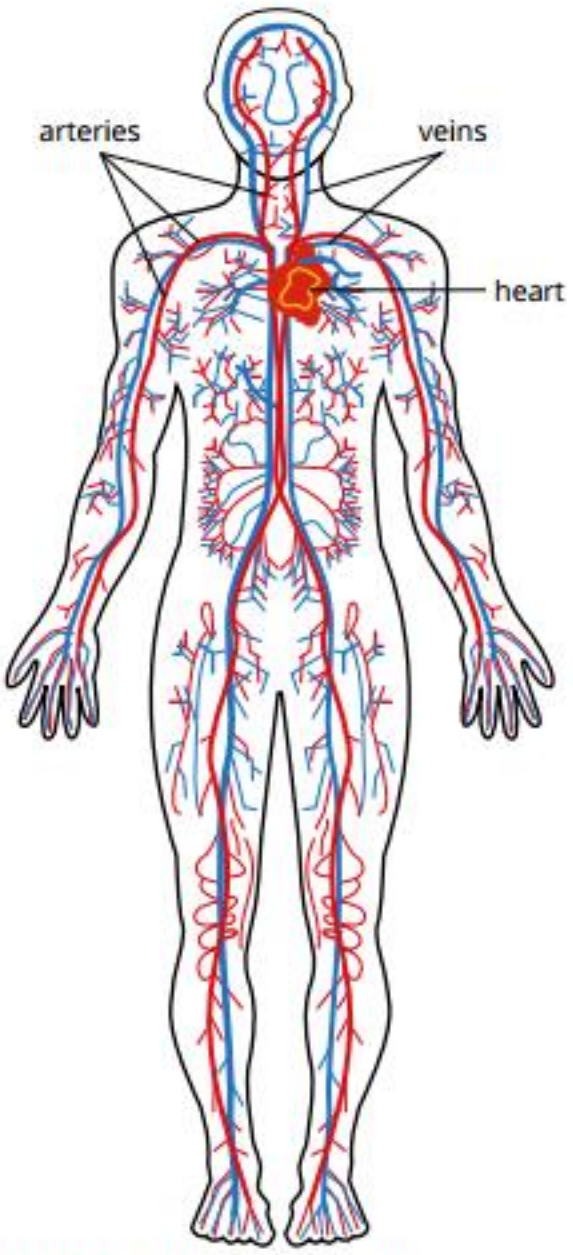


FIGURE 5.1.2 The human circulatory system is an example of a closed circulatory system. The blood in a closed circulatory system is always contained within a system of vessels and a heart.

In humans and all other vertebrates, the circulatory system is a closed system, as shown in Figure 5.1.2. In a closed circulatory system, the blood is enclosed within a system of blood vessels and the heart. In other words, unlike open circulatory systems, blood never leaves the vessels in which it is carried. Typically, a closed system involves:

- a multi-chambered heart
- vessels that carry the blood away from the heart
- specialised vessels called capillaries, which allow the exchange of substances between the blood and the tissues it is servicing
- vessels that return the blood to the heart.

DISTRIBUTING MATERIALS: MAMMALIAN TRANSPORT SYSTEMS

The structure and function of transport systems are similar in all mammals. Mammals have two transport systems: the blood circulatory system and the lymphatic system (Table 5.1.1).

THE CIRCULATORY SYSTEM

The mammalian circulatory system is a closed system that transports substances throughout the body (Figures 5.1.2 and 5.1.3). The vital metabolic products of the body are transported via the blood. The blood and the circulatory tissues and organs ensure that all cells have a ready supply of nutrients and oxygen, and means to transport away metabolic wastes. In mammals, the highly branched network of the circulatory system means that no cell is more than 1 mm from a **capillary**. This ensures there is efficient nourishment and waste removal for all cells in the body.

TABLE 5.1.1 Mammalian transport systems

Blood circulatory system	Lymphatic system
• is a closed circulatory system	• is an open circulatory system
• uses blood as the circulatory fluid	• circulates colourless lymph fluid
• transports the majority of substances to and from cells in mammals	• plays vital roles in maintaining osmotic and fluid balance in tissues and supporting immune defences

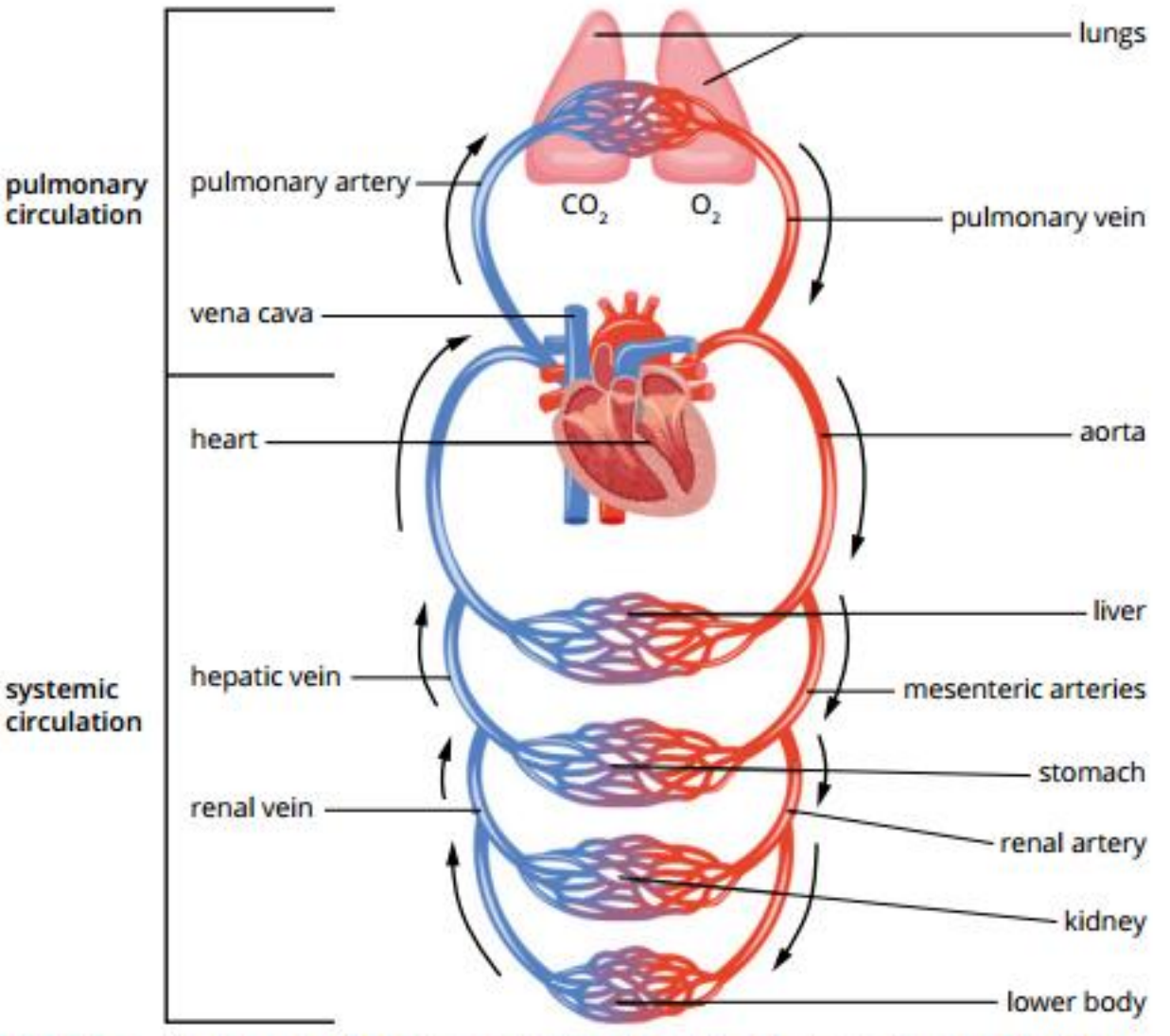


FIGURE 5.1.3 Pulmonary circulation transports blood to and from the lungs. Systemic circulation transports blood to and from all the systems in the mammalian body.

Circulation pathways

Blood circulates around the body via two sequential pathways, as shown in Figure 5.1.3.

- Pulmonary circulation transports blood to and from the lungs. Deoxygenated blood is pumped from the heart to the lungs, where it is oxygenated before returning to the heart.
- Systemic circulation transports blood to and from the rest of the body. This system is larger than the pulmonary circulatory system because the heart must pump blood to all the organs in the body. Oxygenated blood is pumped from the heart to the organs, where it gives up its oxygen to the cells, before returning to the heart.

Components of the circulatory system

The key components of the circulatory system are:

- the heart, which in humans is a four-chambered muscular pump with two pumping chambers (**ventricles**) and two receiving chambers (atria). It is responsible for moving blood throughout the circulatory system. The right side of the heart pumps deoxygenated blood, while the left side pumps oxygenated blood
- veins and arteries, are a network of muscular vessels carrying blood to and from the heart, divided into:
  - pulmonary vessels, which carry blood to and from the lungs
  - systemic vessels, which carry blood to and from all other parts of the body to the heart
  - capillaries, which are numerous very fine vessels with thin walls that provide a large surface area across which exchange of substances occurs, and connects the arteries to the veins
- blood, which is the circulating fluid and is highly specialised for transport and immune defence.

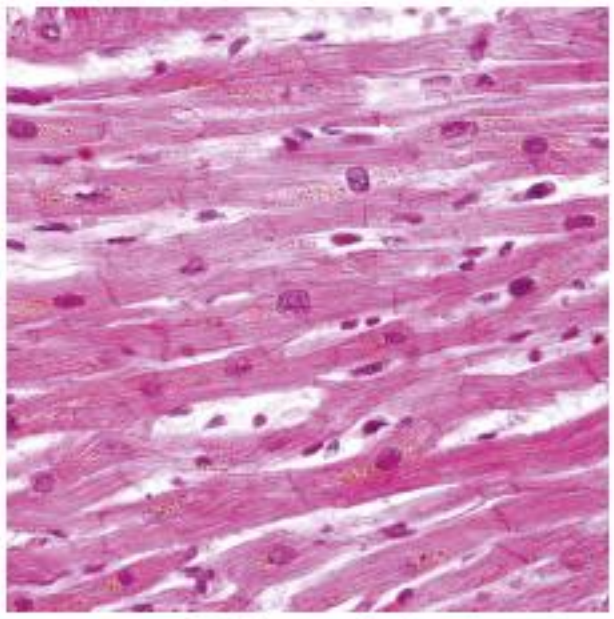


FIGURE 5.1.4 Cardiac muscle tissue is made of highly specialised muscle cells that are found only in the heart. The dark bodies are nuclei.

The heart

The mammalian heart is in the centre of the chest, between the lungs, surrounded by the protective rib cage. It consists of a number of tissues including cardiac muscle (Figure 5.1.4), **connective tissue** and nerve tissue. Connective tissue makes up the **valves**, and nerve tissue controls the heart rate.

The mammalian heart has four chambers, as shown in Figure 5.1.5. The upper receiving chambers, which have thinner walls, are the atria. Each **atrium** opens into one of the lower, thicker-walled chambers, called ventricles. Blood moves through the heart in one direction because of the presence of four one-way valves: one between each atria and ventricle, and one between each ventricle and its outgoing **artery**.

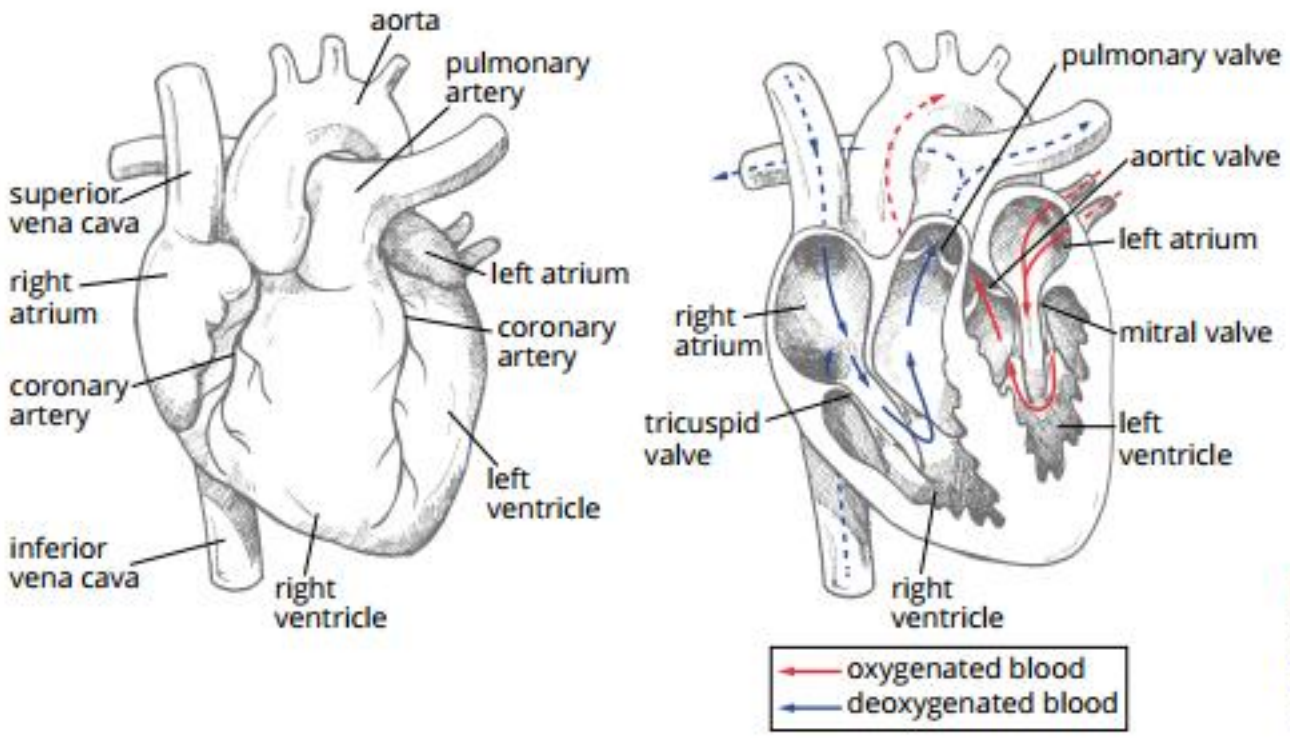


FIGURE 5.1.5 The human heart. (a) View of the ventral surface of the heart. (b) Ventral surface opened to show the major vessels, valves and the greater thickness of the left ventricle

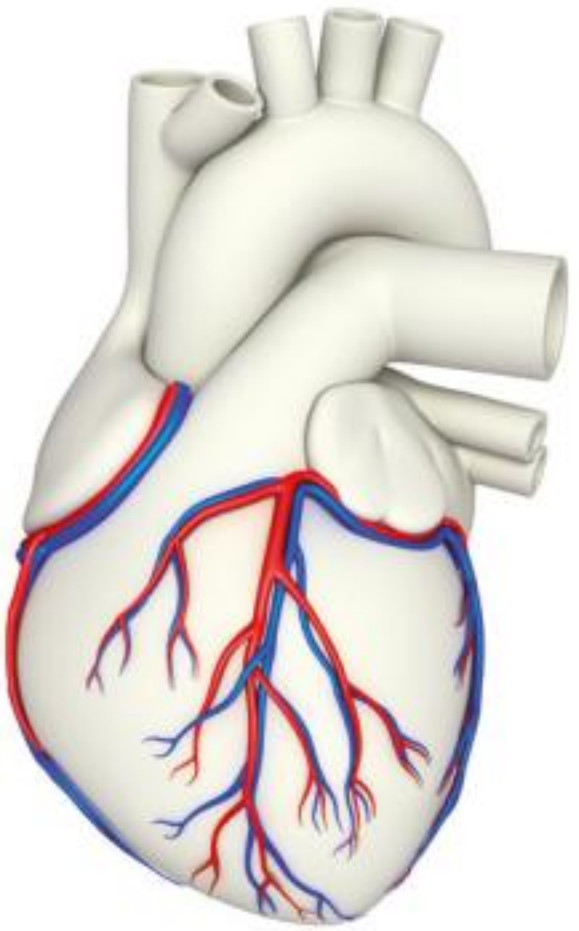


FIGURE 5.1.6 The coronary system, showing the coronary arteries (red) and coronary veins (blue)

Both sides of the heart function in a coordinated way: first, both atria contract; then, both ventricles contract.

Deoxygenated blood (blood with a low oxygen concentration), returns from the body through the venae cavae (singular, vena cava), flowing into the right atrium and through the tricuspid valve into the right ventricle as both chambers relax between contractions. The atrium contracts first, further filling the right ventricle. As the ventricle contracts, the rising ventricular pressure closes the tricuspid valve (between the right atrium and right ventricle) and opens the pulmonary valve (between the right ventricle and the opening of the **pulmonary artery**), causing blood to be ejected into the pulmonary artery towards the lungs.

In the lungs, blood loses carbon dioxide and gains oxygen by diffusion as blood flows through the narrow capillaries around the alveoli. Oxygenated blood (blood with a high oxygen concentration) returns from the lungs to the left atrium through the **pulmonary veins**. The contraction of the atrium forces the blood into the left ventricle. When the left ventricle contracts, oxygenated blood is pumped out of the left ventricle to the rest of the body via the **aorta**. The mitral valve (between left atrium and left ventricle) also closes, preventing backflow into the left atrium.

During a complete circuit around the body, blood passes through each side of the heart. In humans, one complete circuit takes about 45 seconds.

The coronary system

The heart is a continuously active muscular organ, so it has a high requirement for nutrients and oxygen. The cells of the heart have their own rich blood supply via the **coronary circulation**.

The coronary system begins at the base of the aorta and consists of vessels that spread across the surface of the heart and penetrate the heart tissue (Figure 5.1.6).

As the heart is always active, its cells require a constant energy supply. Cells within the heart, therefore, contain many mitochondria to ensure there is always energy available.

Arteries, veins and capillaries

Blood vessels (Figure 5.1.7) are named according to their structure and position in the circulatory system:

- **arteries** (and smaller **arterioles**) transport blood away from the heart
- **veins** (and smaller **venules**) transport blood towards the heart
- **capillaries** are the narrow exchange vessels between arteries and veins.

Arteries and veins are composed of the same layers of tissue, but arteries have more muscular walls to withstand the pressure of the blood as it is pumped out of the heart. Veins are more easily stretched and contain valves, to prevent blood from flowing backwards. Capillaries have very thin walls consisting of only a single layer of flattened epithelial cells.

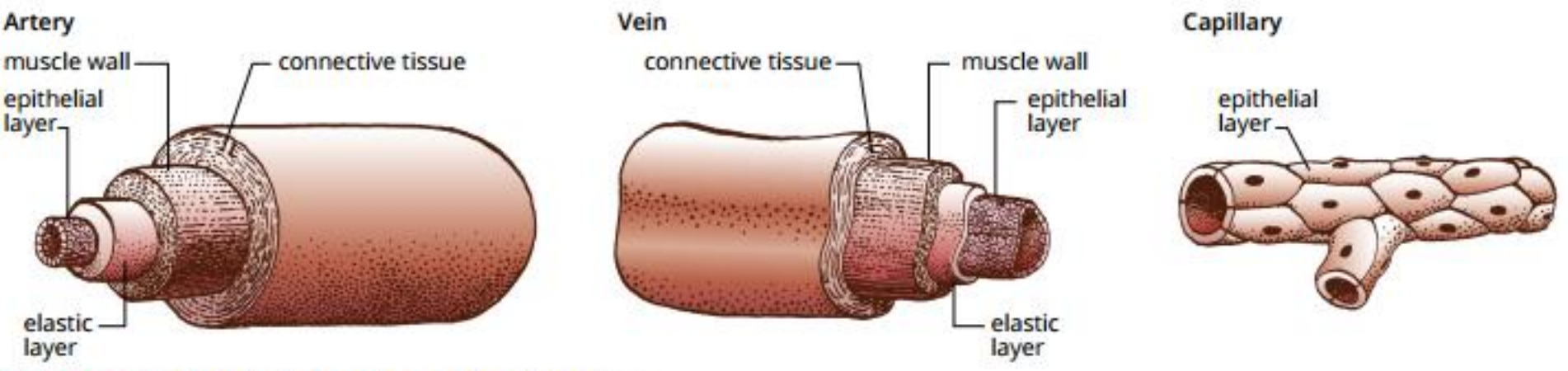


FIGURE 5.1.7 Wall structures of an artery, a vein and a capillary

The structure, function and other features of the blood vessels in the circulatory system are summarised in Table 5.1.2.

TABLE 5.1.2 Structure and function of the blood vessels in the circulatory system

	arteries	veins	capillaries
structure	• consist of an epithelial layer of cells, an elastic layer, muscle wall and connective tissue • same structure as veins but thicker muscular walls	• consist of an epithelial layer of cells, an elastic layer, muscle wall and connective tissue • same structure as arteries but thinner muscle walls	• consist of a single layer of flattened epithelial cells • very thin walls
function	• transport blood away from the heart	• transport blood towards the heart	• connect arteries to veins • deliver nutrients and other substances to extracellular fluids, and receive wastes
other features	• higher blood pressure than veins	• contain many one-way valves • lower blood pressure than arteries	• very numerous • form a network within tissues to be near most cells

Capillaries

Capillaries are the smallest of the blood vessels; their internal diameter is so small that blood cells have to travel through them in a single file (Figure 5.1.8). Their membrane structure makes them very flexible, allowing them to fold and squeeze through the narrow spaces between cells. A capillary has a diameter of 5–10 μm, so red blood cells (about 7–10 μm in diameter) pass very close to the capillary walls. When the wall of a red blood cell presses against a capillary wall, there is an exchange of oxygen and carbon dioxide. The flattened shape and lack of a nucleus in red blood cells are believed to enhance their transport capability by increasing the surface area available for exchange (the importance of surface area-to-volume ratio was covered in Chapter 2).

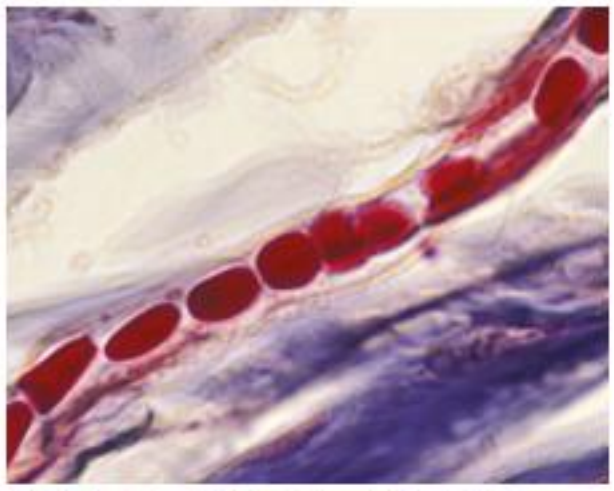


FIGURE 5.1.8 Red blood cells flowing in a single file through a capillary

The capillaries connect arteries to veins, deliver oxygen, nutrients and other substances to extracellular fluids via diffusion, and receive carbon dioxide and other wastes. Capillary walls are extremely thin (just one epithelial cell thick) and porous, which allows substances to pass in and out of the circulatory system into **interstitial spaces**, tissue and cells. Because of their important role in the transport of oxygen and nutrients to tissues, capillaries are most abundant in metabolically active tissues and organs, such as muscle tissue.

Capillaries are distributed throughout the body as an enormous, branched network, providing a vast surface area for the exchange of materials between the blood and extracellular fluid. The interwoven network of blood vessels is known as a capillary bed. To ensure that materials are transported rapidly and efficiently, most cells are no more than 1 mm away from the nearest capillary.

Exchange between blood plasma and extracellular fluid occurs by diffusion and **filtration** across capillary walls. Ions and small molecules such as glucose and amino acids diffuse through the capillary wall, along concentration gradients. Filtration occurs because of two opposite forces: hydrostatic pressure (or blood pressure) and osmotic pressure (Figure 5.1.9). The pressure from these two forces pushes fluid into and out of the capillaries. Hydrostatic pressure is a result of blood pushing outwards on the capillary walls. Osmotic pressure results from the differing solute concentrations between the blood and the extracellular fluid.

i Capillaries are one cell thick, allowing efficient exchange of materials via diffusion and filtration between the blood and interstitial spaces, tissues and cells.

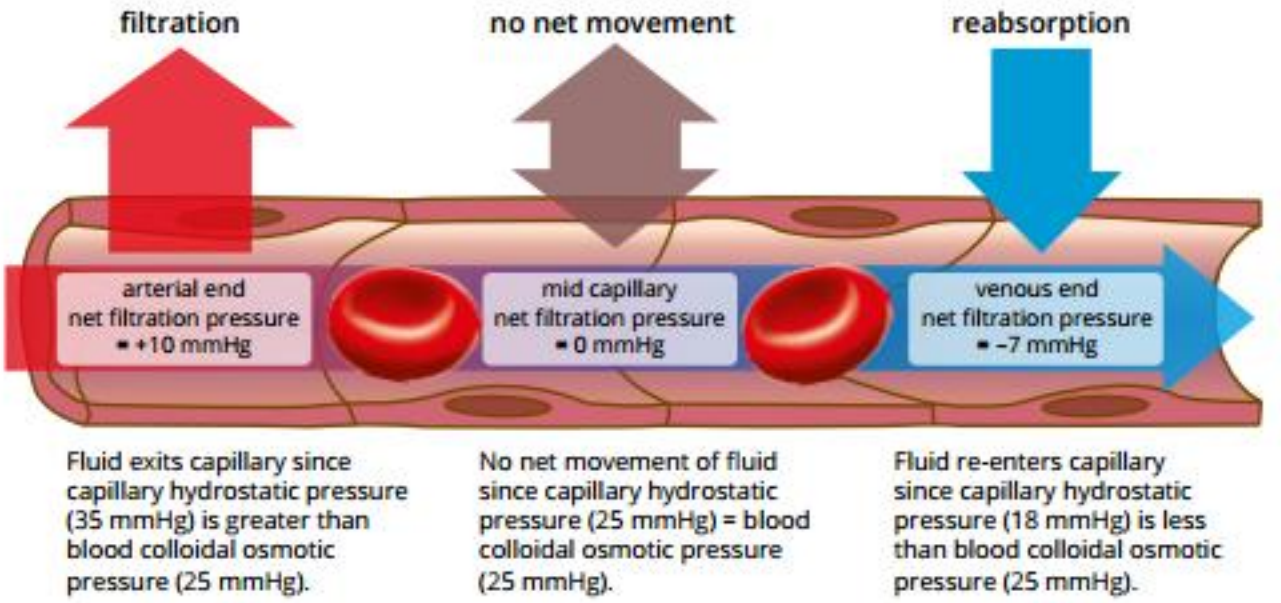

filtration
no net movement
reabsorption
arterial end
net filtration pressure
= +10 mmHg
mid capillary
net filtration pressure
= 0 mmHg
venous end
net filtration pressure
= –7 mmHg
Fluid exits capillary since capillary hydrostatic pressure (35 mmHg) is greater than blood colloidal osmotic pressure (25 mmHg).
No net movement of fluid since capillary hydrostatic pressure (25 mmHg) = blood colloidal osmotic pressure (25 mmHg).
Fluid re-enters capillary since capillary hydrostatic pressure (18 mmHg) is less than blood colloidal osmotic pressure (25 mmHg).

FIGURE 5.1.9 Filtration of fluid across capillary walls is a result of capillary hydrostatic pressure (blood pressure) pushing outwards and blood colloid osmotic pressure pushing inwards. Overall hydrostatic pressure is greater and so fluid leaks into the extracellular environment.

Because blood is hypertonic (more concentrated) than the extracellular fluid of the surrounding cells, water from the cells tries to move through the capillary walls into the blood, putting an inward pressure on the capillaries. The pressure varies along the length of a capillary, but overall, the hydrostatic pressure is greater than the osmotic pressure, so more fluid filters out of the capillary than filters in. This is shown in Figure 5.1.9. This pressure results in a small amount of protein leakage through the capillary wall cells. When blood pressure increases, this leakage is higher and can result in fluid loss to tissues, causing swelling. Reabsorption allows around 85% of the fluid to return to the capillaries, while the remaining 15% enters the lymphatic system.

In some tissues, such as the gut and **liver**, the capillaries are more permeable and allow large molecules to cross. This helps the absorption of digested foods from the gut and enables the liver to take in materials to be broken down. In contrast, capillaries in the brain have very low permeability, and access of substances to brain tissue is tightly controlled. Nerve tissue is very sensitive to its environment, so it is important that the composition of the extracellular fluid surrounding the brain and spinal cord is carefully regulated.

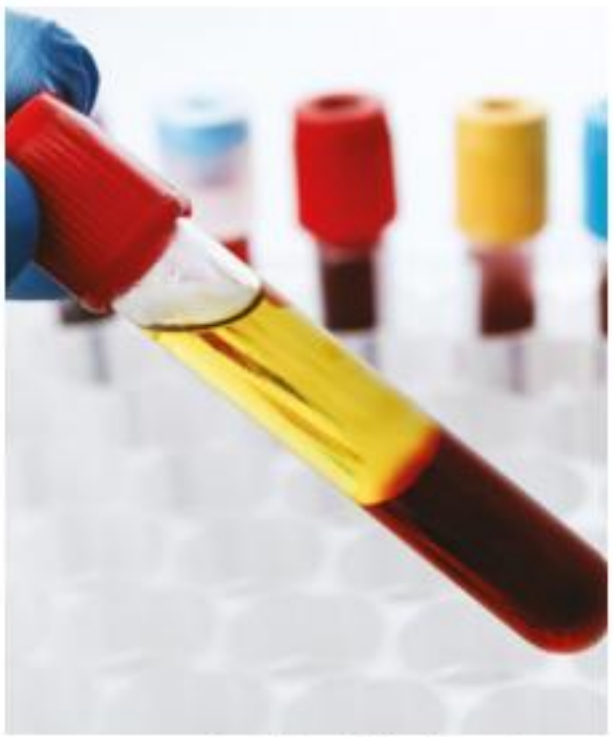


FIGURE 5.1.10 Centrifuged blood separates into plasma (clear yellow fluid) and the cellular elements (dark red solids).

Blood

Mammalian blood is a fluid containing cells and cellular fragments. The fluid portion of blood is plasma, which is a pale-yellow liquid, largely water by volume, and contains ions, dissolved gases, proteins, hormones, nutrients and wastes. The cellular elements of blood include red blood cells (erythrocytes), white blood cells (leukocytes) and platelets, as shown in Figure 5.1.10. They are produced by cells located in the red bone marrow, found in the upper ends of long bones and in flat bones like the skull, ribs and pelvis. Blood is a tissue because it is made up of many similar cells working together.

Red blood cells

In humans, red blood cells make up around 40% of the blood in females and 45% of the blood in males. A single drop of human blood contains about 5 million red blood cells. Mature red blood cells are concave on each side and highly flexible. They lack a nucleus and are full of the protein, **haemoglobin**. Unlike carbon dioxide, oxygen is relatively insoluble. The function of haemoglobin is to bind to the oxygen and transport it to the cells (see Module 5.2). Haemoglobin is also what gives red blood cells their red colour.

White blood cells

White blood cells are slightly larger than red blood cells, but there are far fewer of them (Figure 5.1.11a). A drop of blood contains 5000–10000 white blood cells, but more are held in reserve in organs such as the spleen, kidney, thymus, thyroid gland and lymphatic tissue.

There are several different types of white blood cells within the **immune system**. The two most numerous types are phagocytes (neutrophils and macrophages) and lymphocytes (B cells), both of which are involved in defence against microorganisms (see Chapter 8 for more detail).

- Phagocytes remove debris and fight infections. They are attracted to a site of infection, squeeze through tiny gaps in capillary walls and engulf harmful bacteria and damaged cells.
- Lymphocytes are responsible for the production of antibodies and the development of immune responses.

Platelets

Platelets are fragments of cells. They are much smaller than red and white blood cells and contain substances that are important in preventing blood loss and promoting blood clotting. Platelets are seen in Figure 5.1.11b.

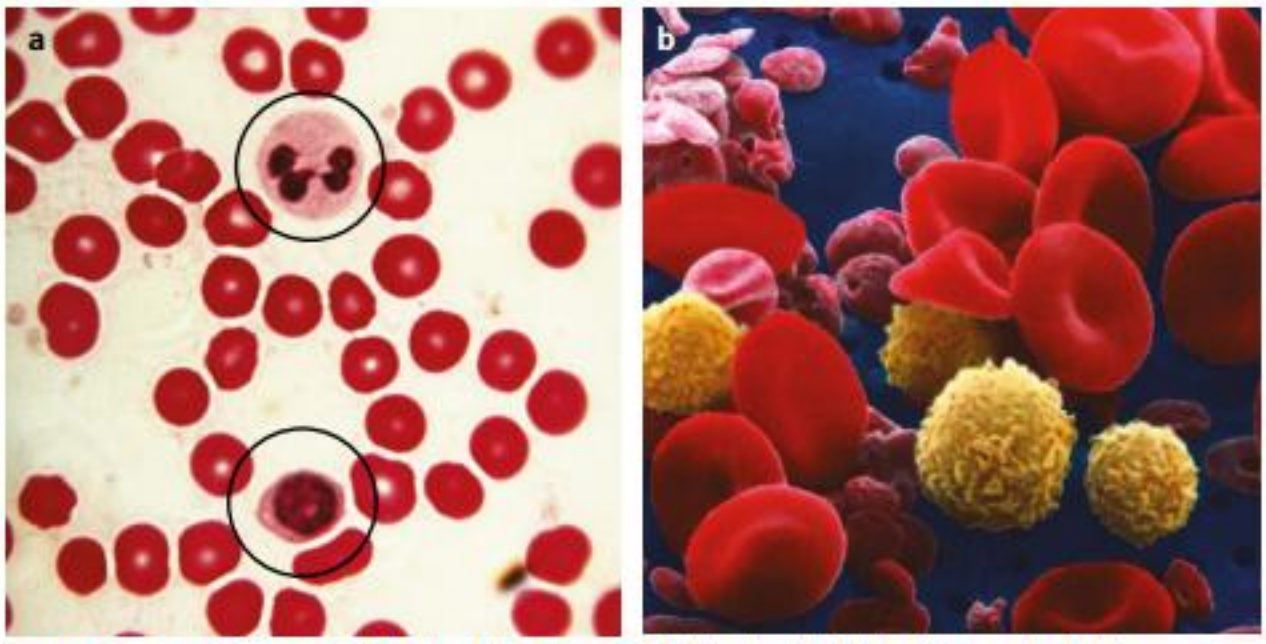


FIGURE 5.1.11 (a) Many red blood cells and two white blood cells (circled) viewed by light microscopy. (b) A coloured scanning electron micrograph showing human blood tissue. The red biconcave discs are red blood cells, the yellow spheres are white blood cells, and the smaller pink cells are platelets.

THE LYMPHATIC SYSTEM

The lymphatic system is the second transport system in mammals. The lymphatic system:

- is an open system
- consists of lymph vessels, lymph nodes and lymphatic organs such as the thymus and spleen
- transports a colourless liquid called lymph
- transports lymph in one direction, from the tissues to the heart.

The lymphatic system has several roles. One of its main roles is to return extracellular fluid containing proteins that have leaked out of the capillaries back into the circulatory system. Without the constant removal of leaked proteins from the extracellular fluid by the lymph capillaries, fluid would accumulate in the tissues. Once inside the lymphatic system, this fluid is called lymph.

The structures of the lymphatic system, especially the lymph capillaries and vessels, is similar to the capillaries and veins of the blood circulatory system (Figure 5.1.12). Fine lymphatic capillaries join to form increasingly larger vessels that eventually empty into the large veins near the heart.

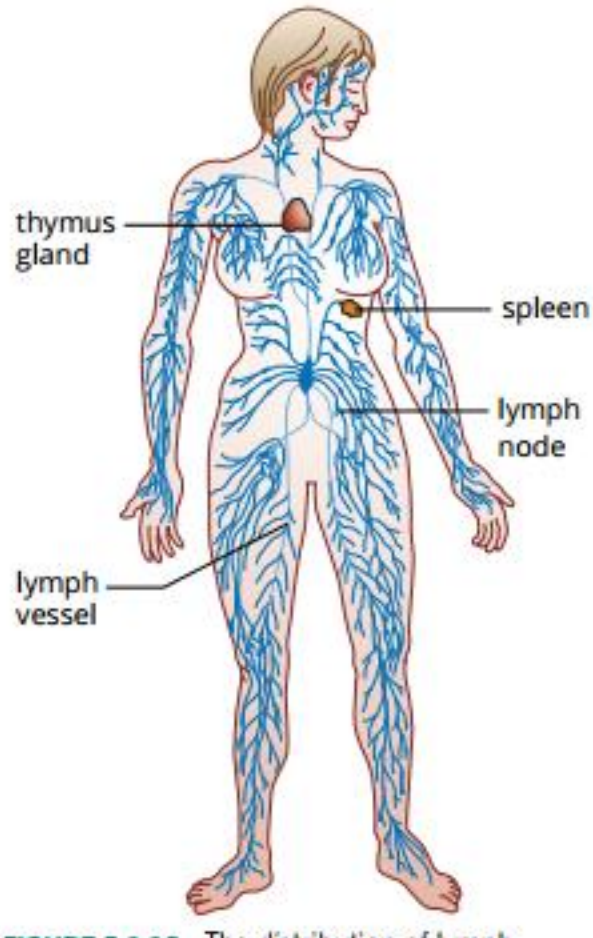


FIGURE 5.1.12 The distribution of lymph vessels throughout the body. The blood circulatory system loses around 3 litres of fluid into the extracellular fluid every 24 hours. The lymphatic system collects and returns this fluid to the circulatory system.

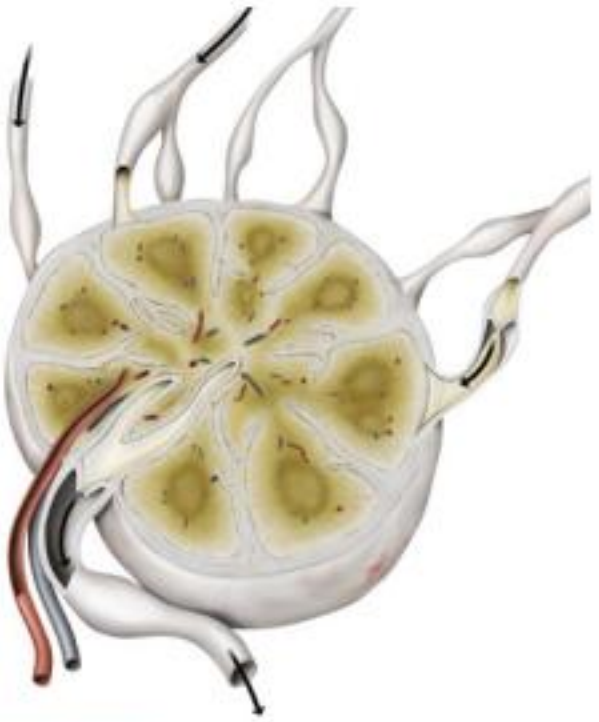


FIGURE 5.1.13 A cross-section of a lymph node. The lymphatic fluid enters the lymph node through lymph vessels where pathogens and cancerous cells are destroyed.

Some of the larger lymph vessels can contract, but most lymph flow results from the external compression of lymph vessels by muscular activity, such as during movement and breathing. When vessels are compressed, the lymph fluid is forced in one direction because of numerous one-way valves, like those in veins, located along the vessels. When a person is inactive (such as standing still or sitting) for a long time, the fluid drainage from tissues decreases and causes swelling. This is especially so in the legs, because fluid drainage must work against gravity.

The lymphatic system also plays a vital role in the immune system. Invading pathogens are transported in the lymph to the lymph nodes (Figures 5.1.12 and 5.1.13), where bacteria, viruses and cancer cells are trapped and destroyed by phagocytes and lymphocytes (see Chapter 8) 320. This is why your lymph nodes swell when you have an infection.

Malfunction of the lymphatic system: deep vein thrombosis

People who sit for long periods of time, such as passengers on a long flight, are encouraged to stretch and exercise regularly to assist the movement of lymph and venous blood back to the heart. Failure to do this can result in swelling in the feet, ankles and legs because fluid accumulates in these areas. This can lead to a condition called deep vein thrombosis (DVT), a blood clot that forms in the veins of the leg. DVT can result in pulmonary embolism, a blockage of the main artery of the lungs, if the clot breaks away and is carried by the bloodstream to a lung and lodges there (Figure 5.1.14). A pulmonary embolism can cause difficulty in breathing, chest pain, heart palpitations, fluid build-up in the lungs and, if the clot completely blocks an artery, death. Regular exercise, a healthy diet and not smoking, reduce the risk of DVT.

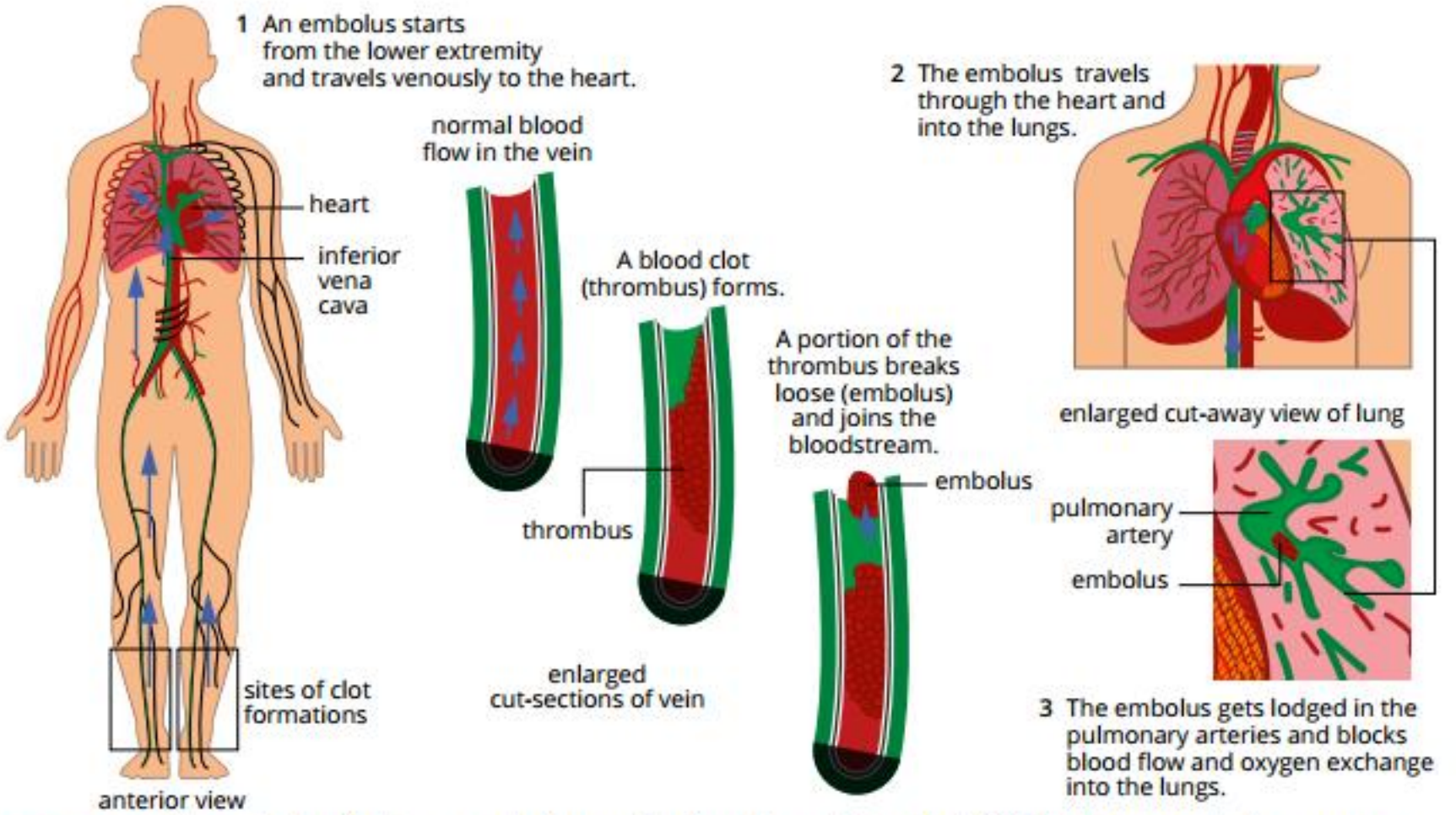

1 An embolus starts from the lower extremity and travels venously to the heart.
heart
inferior vena cava
sites of clot formations
anterior view
normal blood flow in the vein
A blood clot (thrombus) forms.
thrombus
A portion of the thrombus breaks loose (embolus) and joins the bloodstream.
embolus
enlarged cut-sections of vein
2 The embolus travels through the heart and into the lungs.
enlarged cut-away view of lung
pulmonary artery
embolus
3 The embolus gets lodged in the pulmonary arteries and blocks blood flow and oxygen exchange into the lungs.

FIGURE 5.1.14 The mechanism of pulmonary embolism resulting from deep vein thrombosis (DVT). A pulmonary embolism can occur when a blood clot (embolus) breaks off and lodges in a lung, blocking oxygen exchange.

5.1 Review

SUMMARY

- There are two types of circulatory systems in animals: open circulatory systems and closed circulatory systems.
- In closed circulatory systems, the blood is enclosed within a system of blood vessels and the heart.
- Mammals have two transport systems: the blood circulatory system and the lymphatic system.
- The blood circulatory system:
  - consists of the heart, veins, arteries, capillaries and blood
  - capillaries are one cell thick which enable material to diffuse through them, either out of them, for example, oxygen and nutrients, or into them, for example, carbon dioxide and cellular waste
  - transports nutrients and oxygen to all cells in the body and transports metabolic wastes away from all cells in the body
  - has two sequential circulation pathways: pulmonary circulation (transports blood to and from the lungs) and systemic circulation (transports blood to and from the rest of the body).
- The lymphatic system:
  - consists of lymph vessels, lymph nodes and organs, such as the thymus and spleen
  - transports a colourless liquid called lymph
  - transports lymph in one direction, from the tissues to the heart
  - has several functions, one of which is returning extracellular fluid containing proteins that have leaked out of the capillaries back into the circulatory system.

KEY QUESTIONS

Describe

1 Describe the two pathways through which blood is circulated in mammals.

2 List the fluid and cellular components of blood.

3 Name the protein that binds with oxygen in red blood cells.

4 Recall why most exchange of material occurs at capillary level.

5 Identify the systems in the body that are supported by the lymphatic system.

6 Describe the function of the circulatory system.

7 Name the two forces that are responsible for pushing fluid into and out of capillaries. Describe in which direction each of these exert pressure.

Apply

8 Explain the functions of the lymphatic system.

9 Discuss what 'leaky' capillaries are and how they can aid the immune system.

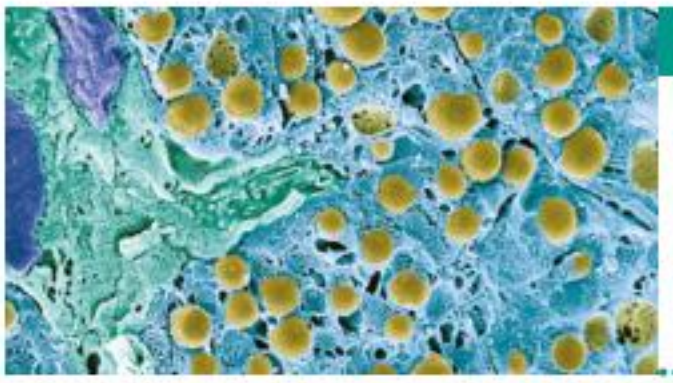
Analyse

10 Compare open and closed circulatory systems.

11 A patient has extremely low blood pressure. Her colloid pressure is normal. Predict, using estimated values, the effect on net filtration pressure through her capillaries.

12 Compare the features of the circulatory system and the lymphatic system.

5.2 Gas exchange in complex animals



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- describe the properties of efficient gas exchange surfaces
- explain the components of the mammalian respiratory system
- explain how complex animals obtain oxygen from their environment
- analyse how the partial pressure of gas influences the rate and direction of gas exchange.

Gas exchange is a fundamental process for all organisms. As you will recall from Chapter 3, all organisms need to exchange oxygen and carbon dioxide with their environment to maintain the important energy-transforming process: cellular respiration. Disruption of this exchange—for example, by respiratory illness in humans—can have serious consequences.

Animals generally have active lifestyles and high energy demands. Their systems are specialised to suit the demands of the environment in which they exist. The respiratory system is responsible for gas exchange in complex animals. It is important to distinguish the organ system of respiration from cellular respiration. Cellular respiration functions to convert nutrients to energy within a cell, while the organ respiratory system in complex animals transports carbon dioxide and oxygen between cells and the external environment.

EFFICIENT GAS EXCHANGE SURFACES

In **aerobic** (oxygen-dependent) organisms, a constant supply of oxygen to cells is required for cellular respiration to occur. Carbon dioxide, which is a waste product of cellular respiration, forms a weak acid in solution with water. If carbon dioxide is allowed to accumulate in body fluids, the pH decreases (i.e. acidity increases), with damaging effects on the structure and function of many important molecules. So, it is important that carbon dioxide is removed efficiently.

In single-celled and very small organisms with high surface area-to-volume ratios, adequate levels of gas exchange occur directly with the environment. In larger animals that have a high metabolic rate and a need for highly efficient gas exchange, well-developed mechanisms to ventilate their gas exchange surfaces are required (Figure 5.2.1). This surface is linked closely to blood transport systems so that gases move efficiently between cells and the environment.

FIGURE 5.2.1 The eastern tube-nosed bat (*Nyctimene robinsoni*) is too large to exchange gases directly with its environment. Like all mammals, it has a complex respiratory system to obtain oxygen from the air and expel carbon dioxide.

Gas exchange always takes place by diffusion across a moist plasma membrane. Diffusion is the passive movement of a substance along its concentration gradient from a region of high concentration to a region of low concentration (see Chapter 2). The immediate environment of cells is the layer of fluid that surrounds them. Even for organisms that get their oxygen from air, oxygen must first dissolve in the layer of extracellular fluid covering the gas exchange surface before it can cross cell membranes and enter the cell or respiratory surface.

Small, uncharged molecules, such as oxygen and carbon dioxide, pass directly through the phospholipid bilayer of the cell membrane. Therefore, they diffuse into or out of cells along their concentration gradient. In contrast to the many nutrients that are actively taken up by organisms, neither oxygen nor carbon dioxide is actively pumped across membranes.

The rate of diffusion of a molecule across a membrane depends on the size and maintenance of the concentration gradient, and on the properties of the membrane itself. The amount of a particular molecule transferred per unit time depends on the membrane's permeability to the molecule, the available surface area of the membrane, and the thickness of the membrane (the distance of diffusion).

For efficient gas exchange:

- the surface area should be as large as possible. This increases the surface area-to-volume ratio, enabling faster rates of diffusion to occur
- the barrier to be crossed (such as cell membranes and fluid layers) should be as thin as possible and should consist of a material that allows the gas to pass through the barrier easily
- there should be an adequate supply of the gas being transferred. If the respiratory surface is not adequately ventilated, the rate of exchange drops
- there should be efficient removal of the substance after transfer. Oxygen is carried away from the respiratory surface, usually by blood. Inadequate blood flow past the respiratory surface allows oxygen to accumulate, so that further transfer is slowed down.

In most large animals, energy is used to ventilate the respiratory surface and to circulate blood past its inner surface. The efficient supply and removal of oxygen maintains a high concentration gradient across the exchange surface, and, therefore, a high rate of diffusion. Energy expenditure is most economical when the rates of **ventilation** and blood flow to the respiratory tissue are matched. For example, when you begin to exercise, you need more oxygen. You breathe more heavily and your heart rate increases. Ventilation and blood flow to the lungs are still matched, but each is at a higher level in order to supply more oxygen.

Ventilation (breathing) is the active movement of the respiratory medium (air or water) across a gas exchange surface.

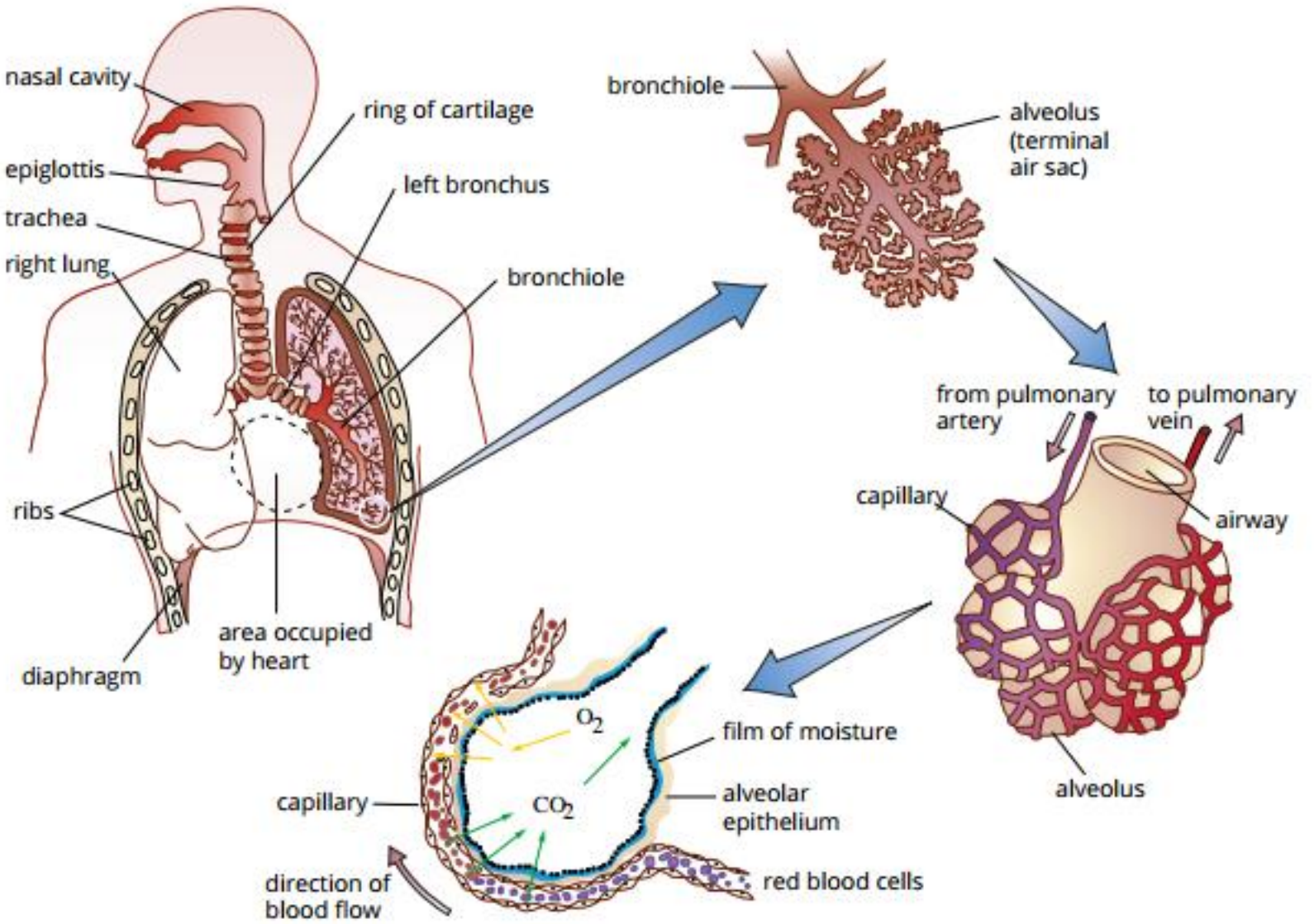


FIGURE 5.2.2 The structure of the human respiratory system

MAMMALIAN RESPIRATORY SYSTEMS

All mammals obtain their oxygen by breathing air. Oxygen is absorbed from the environment by the respiratory system and is then transferred to cells via the circulatory system.

The key steps in the process of mammalian respiration occur at the following sites, shown in Figure 5.2.2:

- Nose and throat—air is drawn in through the nasal cavity and passes into the **pharynx** (the back of the throat). Breathing through the nose is preferable to breathing through the mouth because the air is filtered, moistened and warmed in the nasal passages.
- Airways—from the pharynx, air passes into the airways: the **trachea**, paired **bronchi** and branching bronchioles. The trachea and bronchi are lined with cells covered in **cilia** and secrete mucus (Figure 5.2.3). Particles of dust and bacteria are trapped by this mucus and swept by the cilia back up to the pharynx and swallowed. The **larynx**, containing the vocal cords responsible for speech, is located at the upper end of the trachea.
- **Alveoli**—air enters the terminal **air sacs**, called alveoli, where gaseous exchange takes place. A constant supply of oxygen to cells is the most critical input for endotherms, such as mammals and birds, because they use energy to warm their bodies, and therefore need oxygen at a great rate for cellular respiration.

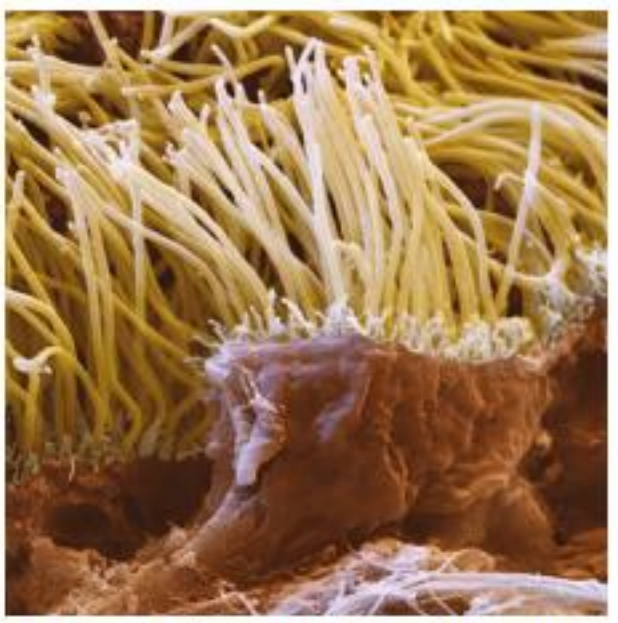


FIGURE 5.2.3 A scanning electron microscope image of a cross-section through the cilia-covered epithelial cells of the bronchus (lung airway). The cilia are microscopic hairs that sweep trapped particles and mucus away from the gas-exchanging parts of the lungs and towards the throat, where the particles can be swallowed or coughed up.

The tissue of the alveoli shows all the features of efficient exchange structures. It provides a large surface area for gas exchange; the total surface area in most adults is 30–70 m^2. Each alveolus is lined with a very thin layer of flattened cells, called the alveolar **epithelium** (Figure 5.2.3). This thin layer of cells is richly supplied with blood through numerous capillaries, facilitating diffusion of gases between the alveoli and the capillaries (Figure 5.2.2). Once oxygen enters the capillaries, it has entered the circulatory system, and the oxygenated blood is transported throughout the body.

Organisms can obtain oxygen either by breathing air (e.g. mammals) or from water (e.g. fish). However, there are two great advantages of obtaining oxygen from air. First, ventilation with air requires less energy than breathing in water, which is heavy.

Second, much more oxygen is available in air than in water. But animals that breathe air must have a large, moist gas exchange surface. The disadvantage of breathing air is that water evaporates continuously from this large, moist surface area. Respiratory surfaces are therefore a major site of water loss for all terrestrial organisms.

Enclosing the respiratory surface inside the body provides physical protection from the external environment, support for the respiratory membrane, and reduces water loss. However, it means that specialised systems must aid in the ventilation of gas to and from the enclosed gas exchange surface.

Lung ventilation

Mammalian lungs are contained in the chest cavity (the thorax), which is completely enclosed and under a small negative pressure that keeps the lungs expanded within the thorax. The floor of the chest cavity is the muscular diaphragm.

Mammals use a 'suction pump' mechanism to ventilate their lungs. The chest cavity is expanded by contracting the diaphragm downwards and raising the ribs. This expands the lungs and draws air in through the airways (Figure 5.2.4). **Inhalation** is always an active process (it requires energy). However, **exhalation** is normally the result of the elastic recoil of the thorax as it returns to its relaxed state. Forceful exhalation involves an active compression of the rib cage.

Tidal volume

Tidal volume is the volume of air moved in and out at each breath. Normal resting levels of inhalation and exhalation are much less than our **vital capacity**, which is the maximum volume of air that we can move into and out of our lungs. Tidal volume varies according to the need for oxygen.

Air moves tidally into and out of mammalian lungs through the same airways. This is not as efficient as one-way flow, because at the end of an exhalation there is still some 'stale' air left in the airways and in the alveoli. The next inhalation draws this stale air back into the lungs, so it is impossible to fill our lungs completely with fresh air. The volume of air left in the respiratory system at the end of exhalation is referred to as the residual volume. Figure 5.2.5 graphically represents lung ventilation in humans.

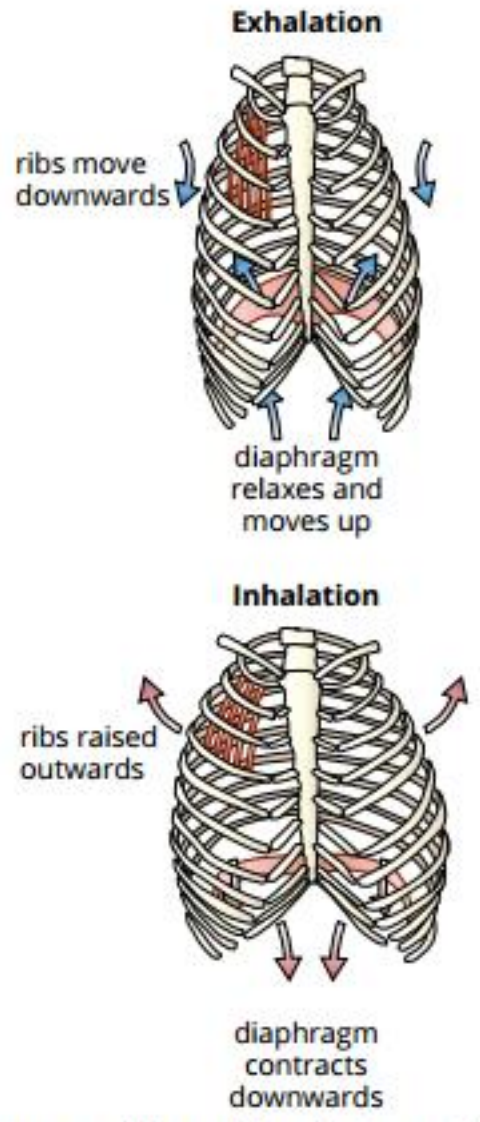


FIGURE 5.2.4 Mammals breathe by negative pressure ventilation. Raising the ribs and contracting the diaphragm increases the volume of the chest cavity and draws air into the lungs (inhalation). Relaxing these muscles causes the volume of the chest cavity to reduce and air is forced out again (exhalation).

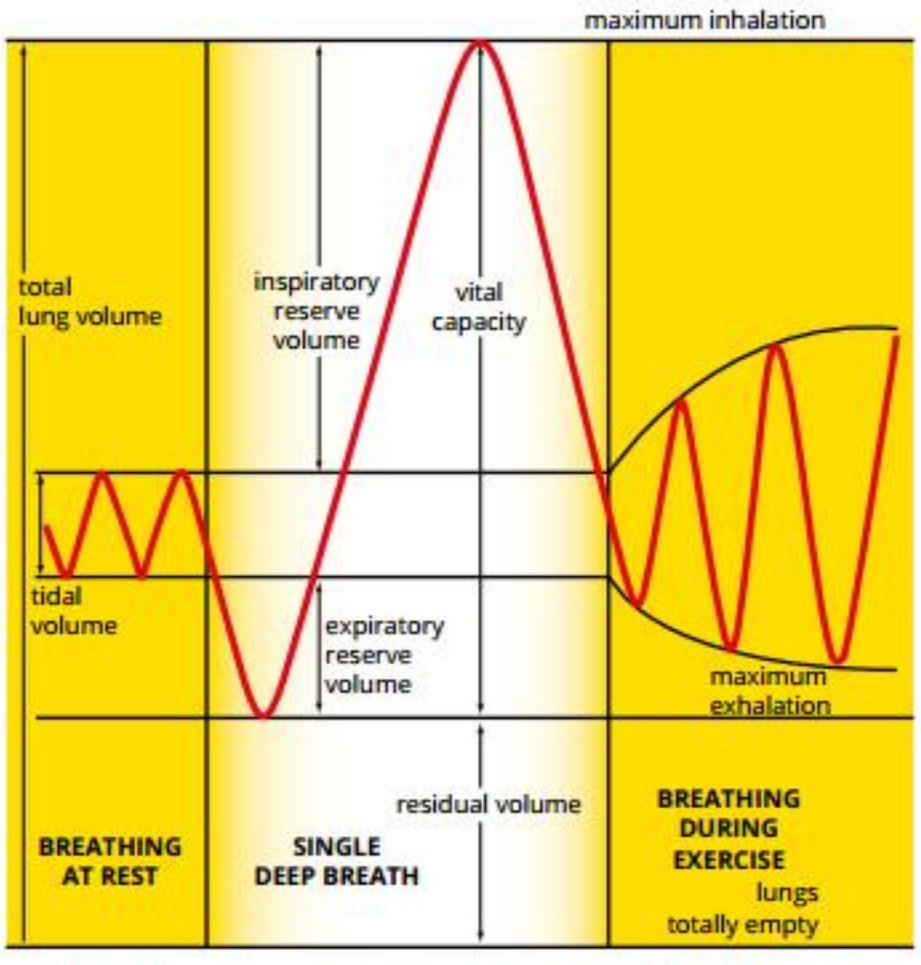


FIGURE 5.2.5 Lung ventilation in humans. During exercise, tidal volume increases. The extent of this increase is directly related to the increased use of oxygen and is matched by an increased blood flow to the lungs.

EXCHANGING AND TRANSPORTING GASES

The exchange and transporting of the two gases, oxygen and carbon dioxide, is possible because of the coordination between the circulatory and respiratory systems. Gas exchange always occurs in the capillaries, whether at the alveoli or in body tissue. Both gases are also transported by the blood.

Gas exchange

Gas exchange, whether in the alveoli or tissues, occurs by diffusion. The rate of diffusion is influenced by the **partial pressure** of gases at the sites of gas exchange. Partial pressure is the pressure a particular gas contributes to the total pressure of a gas mixture. The partial pressure of gas X is written as P_X.

For example, air is a gas mixture that is approximately made up of 78% nitrogen, 21% oxygen and 1% other gases. At sea level, the pressure of air is 760 mmHg.

The partial pressure of the gases that make up air at sea level is:

$P_N = 78\%$ of $760\,\text{mmHg} = 592.8\,\text{mmHg}$

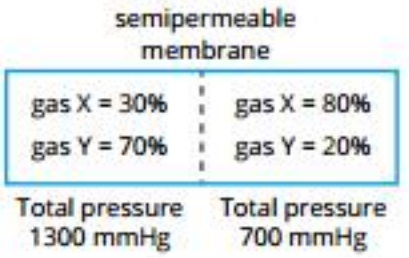
$P_{O_2} = 21\%$ of $760\,\text{mmHg} = 159.6\,\text{mmHg}$

$P_{\text{other gases}} = 1\%$ of $760\,\text{mmHg} = 7.6\,\text{mmHg}$

Worked example 5.2.1

CALCULATING PARTIAL PRESSURE

Consider the following diagram. Calculate the partial pressure of the gases in each side of the container to determine in which direction the gases diffuse.

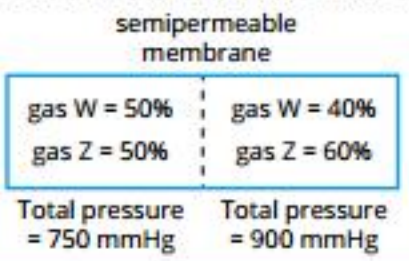


Thinking	Working
Calculate the partial pressure of the gases in the left side of the container.	$P_{X\text{left}} = 30\%$ of $1300 = 390\,\text{mmHg}$ $P_{Y\text{left}} = 70\%$ of $1300 = 910\,\text{mmHg}$
Calculate the partial pressure of the gases in the right side of the container.	$P_{X\text{right}} = 80\%$ of $700 = 560\,\text{mmHg}$ $P_{Y\text{right}} = 20\%$ of $700 = 140\,\text{mmHg}$
Compare partial pressures of gas X. Recall that gases move down partial pressure gradients.	$P_{X\text{left}} = 390\,\text{mmHg}$, $P_{X\text{right}} = 560\,\text{mmHg}$ Gas X moves from right to left.
Compare partial pressures of gas Y. Recall that gases move down partial pressure gradients.	$P_{Y\text{left}} = 910\,\text{mmHg}$, $P_{Y\text{right}} = 140\,\text{mmHg}$ Gas Y moves from left to right.
Answer	Gas X moves from right to left, while gas Y moves from left to right.

➤ Try yourself 5.2.1

CALCULATING PARTIAL PRESSURE

Consider the following diagram. Calculate the partial pressure of the gases in each side of the container to determine in which direction the gases diffuse.



Gas partial pressure in the human circulatory system

As air enters the lungs, water vapour is added to it and oxygen is continually taken up by the body, while carbon dioxide is added. By the time inhaled air reaches the alveolus, the partial pressure of oxygen is 104 mmHg and the partial pressure of carbon dioxide is 40 mmHg.

Gas exchange occurs because gases diffuse down partial pressure gradients.

In the deoxygenated blood in the pulmonary arteries, the partial pressure of oxygen is 40 mmHg and carbon dioxide is 45 mmHg. Consequently, oxygen moves down its partial pressure gradient from the alveoli into the blood. Carbon dioxide also moves down its concentration gradient; it moves from the deoxygenated blood into the alveoli. After gas exchange at the alveoli, the partial pressures of gases in the oxygenated blood leaving the lungs are: $P_{O_2} = 104$ mmHg, $P_{CO_2} = 40$ mmHg.

The cells in your body use oxygen for cellular respiration and produce carbon dioxide as waste. The partial pressures of gases in tissue cells are: $P_{O_2} = 20$ mmHg, $P_{CO_2} = 46$ mmHg. Again, because of the differences in gas partial pressures, oxygen diffuses into the tissue cells while carbon dioxide diffuses into the blood. The deoxygenated blood then returns to the right side of the heart and then to the lungs, via the pulmonary system.

Figure 5.2.6 shows the partial pressure of oxygen and carbon dioxide at the sites of gas exchange and in the blood in the body. The direction in which the gases diffuse is also shown.

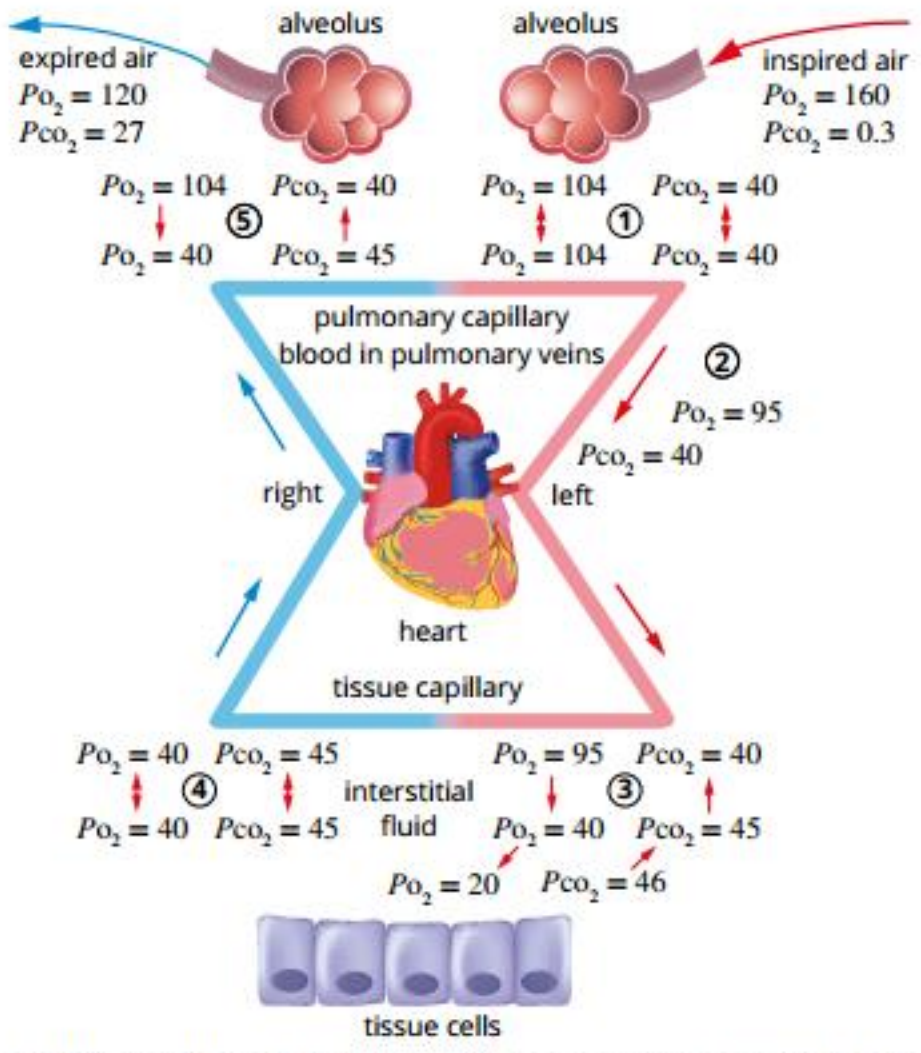


FIGURE 5.2.6 Partial pressure of gases (mmHg) in the respiratory and circulatory systems. Gas exchange occurs down partial pressure gradients.

Carrying oxygen

Maintaining an oxygen concentration gradient across the lung surface requires efficient supply (through ventilation) and removal (by circulation) of the oxygen. But the amount of oxygen that dissolves in water (or blood, which is about 90% water) is very small.

The oxygen-carrying molecule haemoglobin increases the **oxygen-carrying capacity** of the blood; that is, the amount of oxygen that it can carry. The most important feature of haemoglobin is that it can combine reversibly with oxygen.

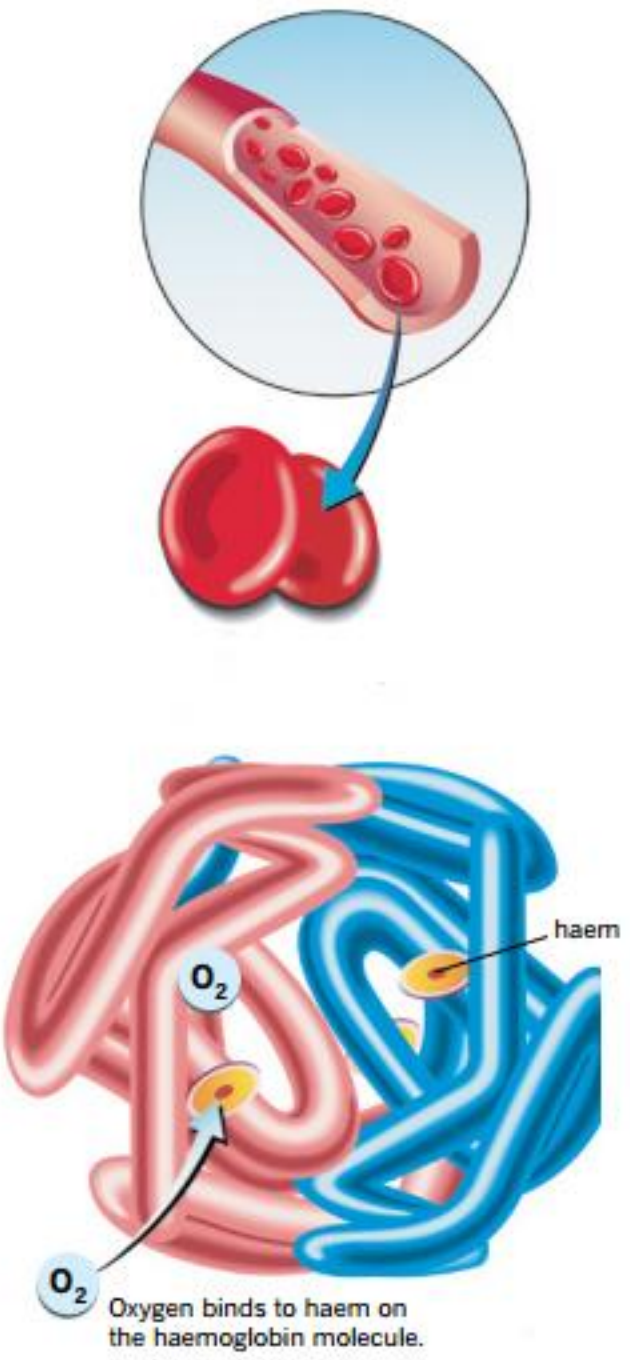


FIGURE 5.2.7 The 3D structure of a haemoglobin molecule. This molecule is responsible for binding oxygen molecules in red blood cells.

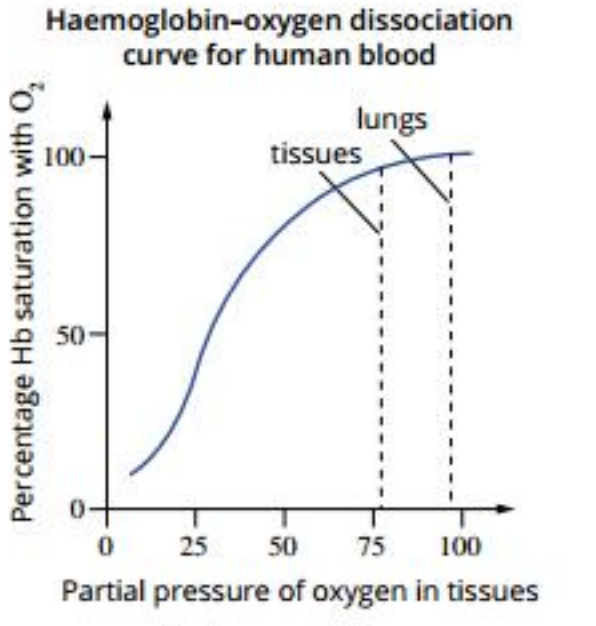


FIGURE 5.2.8 The haemoglobin–oxygen dissociation curve for human blood. It shows how the partial pressure of oxygen effects the extent to which oxygen binds to haemoglobin (Hb) molecules in the body.

Haemoglobin increases the oxygen-carrying capacity of blood and reduces the amount of energy that must be spent pumping blood. Because haemoglobin enables each millilitre of blood to carry much more oxygen, an animal can have a much smaller volume of blood, and pump it around the body more slowly, while still supplying the same amount of oxygen to its cells.

Haemoglobin

Oxygen is relatively insoluble in blood: only 0.2 mL of oxygen gas dissolves in 100 mL of blood. The carrying capacity of mammalian blood is increased 100 times by the presence of the red respiratory protein haemoglobin, which is carried in red blood cells.

Haemoglobin is a complex protein containing iron. Four oxygen molecules can combine with each haemoglobin molecule (Figure 5.2.7). In areas of high oxygen concentration, such as in the blood in vessels in the lungs, haemoglobin combines with oxygen to form **oxyhaemoglobin**. In areas of low oxygen concentration, such as in exercising muscles, oxygen is released (dissociated) from the oxyhaemoglobin. Therefore, the percentage of oxygen concentration in exercising muscles, tissues and lungs varies. This relationship can be seen in Figure 5.2.8.

Oxygen in the tissues

In resting humans, haemoglobin is almost 100% saturated with oxygen in the lungs and about 75% saturated in tissues of other organs (Figure 5.2.8). In the lungs, the high partial pressure of oxygen means that haemoglobin has a high affinity to oxygen and is 98% saturated. However, in the tissues of other organs, the partial pressure of oxygen is around 40 mmHg. At this pressure, haemoglobin has a lower affinity for oxygen. This means that oxygen is released from the haemoglobin molecule and diffuses into cells in the body tissues.

Our muscles are red because they also contain a form of haemoglobin called **myoglobin**. Myoglobin carries a reserve store of oxygen that muscles can use for a limited period if the amount of oxygen in the blood suddenly decreases to a very low level. This situation could arise if a blood vessel were temporarily blocked, or during strenuous exercise. When blood supply is restored, the myoglobin oxygen store is immediately refilled from the blood. Myoglobin has a higher affinity for oxygen than haemoglobin and therefore can take oxygen from it. This also means that haemoglobin releases large amounts of its bound oxygen to exercising muscle before the myoglobin releases its store, making it an emergency resource.

Anaemia

Anaemia is a condition in which there are insufficient red blood cells, or the quality of the red blood cells or the haemoglobin is low. The most common cause of anaemia is a deficiency of iron in the diet. Other causes include failure to absorb iron because of disease, heavy menstruation and inherited disorders such as sickle-cell anaemia.

People with anaemia may have a normal blood oxygen saturation reading, but because they have less functioning haemoglobin in their blood, less oxygen may be getting to the cells in their body. Consequently, anaemia results in pale skin, tiredness, muscle weakness, headaches and problems with concentration. Treatment is determined by the underlying cause, but can involve a change in diet, iron supplements, surgery, or (in extreme cases) oxygen therapy and blood transfusions.

Carrying carbon dioxide

Carbon dioxide, produced by cellular respiration, must be carried in body fluids to an external surface where it can be released to the environment. Because carbon dioxide combines with water to form an acid (carbonic acid) and causes a decrease in pH, it can be carried in solution only in limited amounts.

In mammals, about 5–7% of the carbon dioxide carried by blood is dissolved in the plasma. About 23% combines with haemoglobin molecules (forming carbamino-haemoglobin), but at a different site on the haemoglobin molecule to the site where oxygen binds. Carbamino-haemoglobin is still able to combine with oxygen.

The remainder of the carbon dioxide produced in working tissues passes into red blood cells, where it is converted to hydrogen carbonate ions, and then passes out to be transported in the plasma.

When the blood reaches the lungs, the hydrogen carbonate moves back into the red blood cells where it is converted to carbon dioxide for release during breathing.

5.2 Review

SUMMARY

- Gas exchange is a fundamental process for all organisms.
- Single celled organisms, like bacteria, can rely on diffusion as the only mechanism of gas exchange
- Complex animals, like mammals, have respiratory systems to ensure adequate ventilation.
- Gas exchange always takes place by diffusion across a moist plasma membrane.
- Features of an efficient gas exchange surface are:
  - a large surface area-to-volume ratio
  - moist
  - one to two cells thick
  - adequate supply of the gas being transferred
  - efficient removal of the substance after transfer.
- Mechanisms of gas exchange reflect the environment in which the organisms are found.

KEY QUESTIONS

Describe

1 Name the fundamental process that all organisms rely upon for gas exchange.

2 Identify the advantages of breathing air over water.

3 State the name given to the volume of air that is moved in and out at each breath.

4 List the features that are required for surfaces to be efficient at gas exchange.

5 Recall how most carbon dioxide is moved into the lungs.

6 Describe the concept 'partial pressure of a gas'.

Apply

7 Explain why it is important for respiratory surfaces and cells within the body of animals to be surrounded by extensive capillary systems.

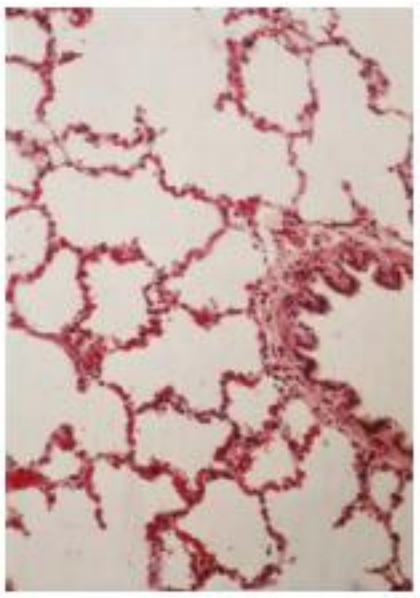
8 Discuss the different mechanisms within humans that can aid in the efficient working of the lungs.

Analyse

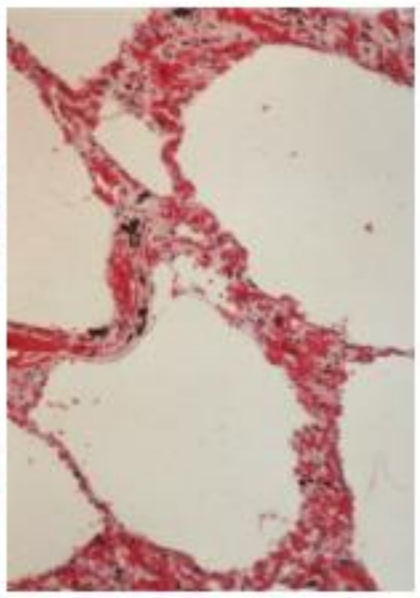
9 Differentiate between cellular respiration and respiration that occurs in a respiratory system.

10 Pneumonia is an infection of the lungs where the alveoli fill with fluid or pus. Predict how pneumonia would impact the efficiency of gas exchange.

11 Emphysema causes the breakdown of alveoli. The following images show the difference between lung tissue in a normal person and a person with emphysema. Deduce how emphysema reduces the efficiency of gas exchange in a diseased person.



Normal lung tissue has large air spaces.

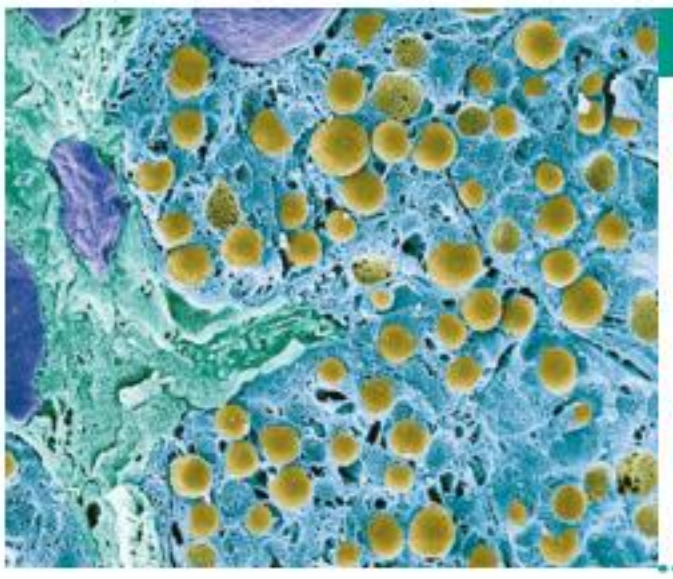


In emphysema, the air sacs break down.

12 A gas mixture comprises 40% oxygen, 25% carbon dioxide and 35% nitrogen. Calculate the partial pressure of the three gases if the total gas pressure is 1250 mmHg.

13 Distinguish between the properties of haemoglobin and myoglobin and consider the role each plays within the body.

5.3 Exchange of nutrients and wastes in complex animals



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- describe the structure and function of the organic molecules that are the products of digestion
- describe the role of the various organs and enzymes used within the mammalian digestive system
- identify foregut and hindgut fermenters
- explain how food is digested by both physical and chemical means
- identify the nephron as the functional unit of the mammalian kidney
- explain the three main stages of urine formation in mammals: filtration, reabsorption and secretion.

Mammals are heterotrophs; unlike plants, they cannot make organic molecules from inorganic materials. Consequently, they must consume other organisms or their products to obtain organic molecules. As well as needing organic molecules to provide chemical energy, heterotrophs also require other organic molecules such as vitamins, amino acids and fatty acids. Their diet must also contain minerals and water.

DIGESTION OF FOOD

Organisms are composed of many different types of complex organic molecules. When eaten as food, these molecules are too large to be simply absorbed into an animal's body. Regardless of the type of animal, food molecules must be small enough to pass across plasma membranes into the cells lining the gut. This is the purpose of digestion—to rapidly break down organic food into molecules small enough to be able to pass through membranes and into cells.

The food you eat does not become part of your body until it has been absorbed by the cells lining the walls of your intestine. The digested food then passes into the bloodstream and is carried throughout the body. If food is not absorbed, it continues through the intestine and is passed out again as faeces (**egestion**).

Digestion is the breakdown of food into a form that can be used by an organism for **metabolism**. This involves physical and chemical breakdown.

Do not confuse egestion with **excretion**. Excretion refers to the removal of substances that were once part of the body and occurs largely in the kidneys.

Physical breakdown

Enzymes are biological catalysts (see Chapter 3). **Digestive enzymes** can only act on the outside surface of food. If food is swallowed in large pieces, the enzymes have a relatively small surface area to work on. Unless the digestive system is extraordinarily long, most of the food would remain undigested. Given the relationship between surface area and volume (see Module 2.4), digestion is much faster if food is in small pieces and the enzymes have a proportionally larger area to act upon.

Therefore, it is important to have a mechanism for breaking down large food into smaller pieces to increase its surface area. Animals have developed a variety of structures to break down food physically. For example, the teeth of vertebrates break food into pieces small enough to be swallowed (Figure 5.3.1). Other organisms, such as birds, reptiles and fish, use the gizzard, an organ in their digestive system, to digest food.

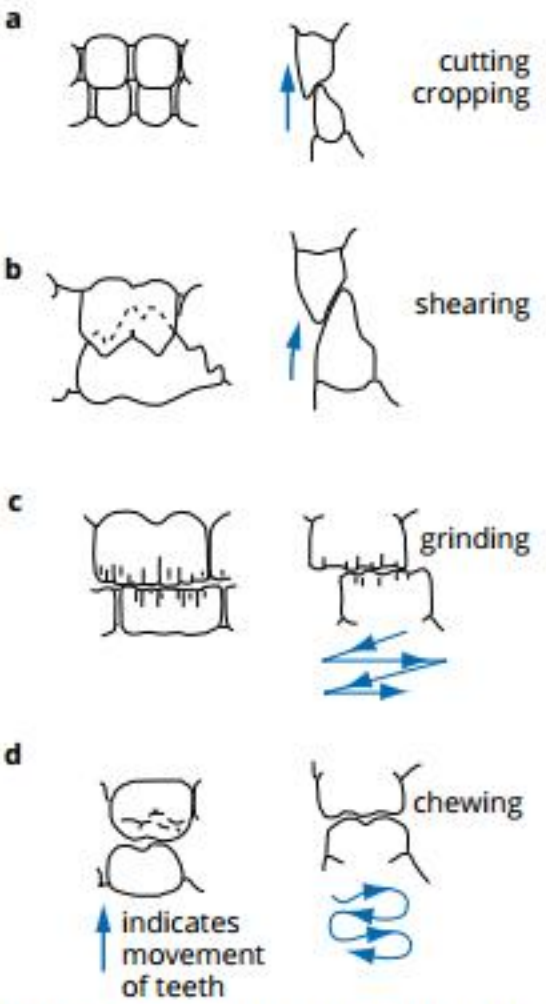


FIGURE 5.3.1 In mammals, the tooth structure is adapted for the mechanical breakdown of different types of foods. (a) Incisors are typically used for cutting and tearing. (b) Carnivores have large powerful cheek teeth that shear through tough tissues and bones. (c) Herbivores have molars that grind fibrous plant foods. (d) Omnivores, such as you, have molars that roll and crush a variety of foods.

Sometimes hard objects, such as stones, are swallowed to assist the grinding process in gizzards (Figure 5.3.2).

To improve the efficiency of digestion, this physical breakdown should take place before **chemical digestion** is completed. In contrast to chemical digestion, physical breakdown does not chemically change food molecules.

Bile is important in the physical breakdown of fats, but it is not an enzyme. Bile is produced by the liver and released into the small intestine where it acts like a detergent to emulsify fats—breaking up large fatty masses into small droplets. This increases the surface area of fats available for digestion by lipases.

FIGURE 5.3.2 Emus (*Dromaius novaehollandiae*) swallow stones, which stay in the gizzard, to aid physical digestion of food.

Chemical digestion

The process of breaking apart complex molecules into simple molecules is called chemical digestion; it is carried out by the action of enzymes. Enzymes are important in digestion because they greatly increase the rate of breakdown of food molecules.

Most digestive enzymes split food molecules by the process of **hydrolysis** (from Greek *hydro*, meaning 'water', and *lysis*, meaning 'to split'). This means they split the food molecule at a particular point by adding a water molecule. There are three main kinds of digestive enzymes, as shown in Figure 5.3.3. These enzymes split biomacromolecules into their constituent building blocks, which can then be used by the body.

- **Amylases**, which act on carbohydrates to release simple sugars.
- **Proteases**, which act on proteins to release amino acids.
- **Lipases**, which act on lipids to release fatty acids and glycerol.

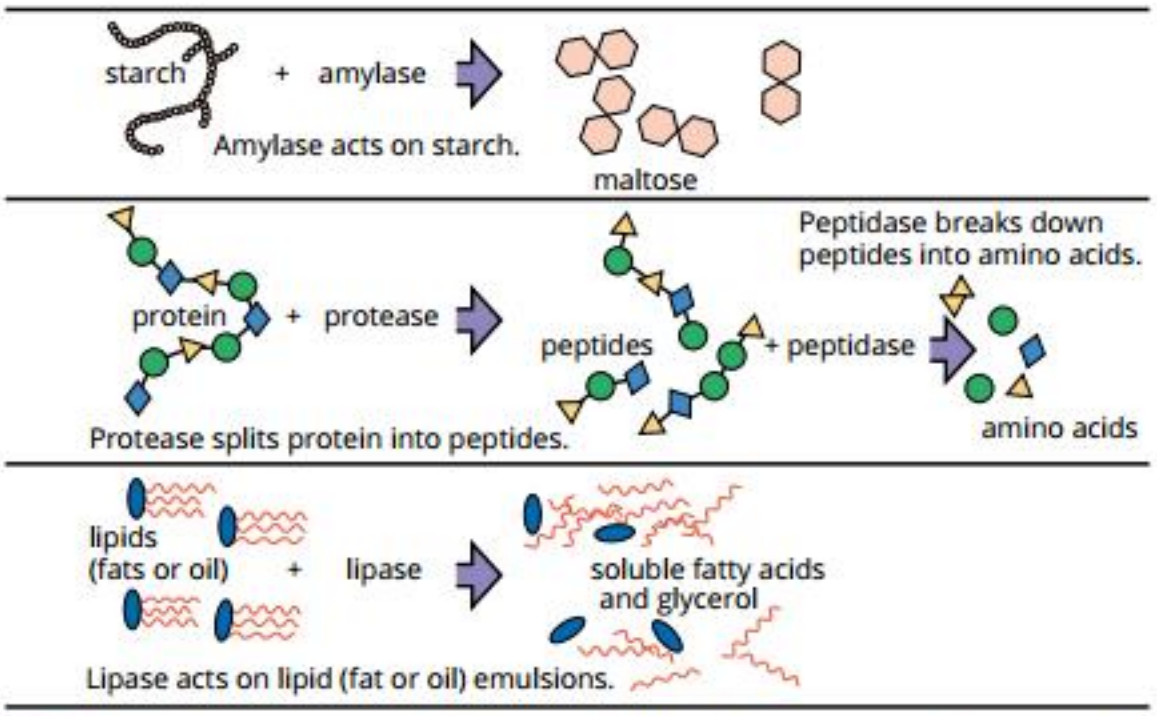


FIGURE 5.3.3 Digestion involves the splitting of food molecules into components small enough to pass across plasma membranes and into the body. Enzymes used in digestion are often named according to the substance on which they act, with the common ending -ase. For example, protease digests proteins and lipase digests lipids.

Humans secrete more than 20 different digestive enzymes into the digestive tract. Digestive enzymes are manufactured by specific cells in the gut wall, and by the salivary glands and the pancreas. Many very large food molecules can be broken down only by several enzymes acting one after the other. In this case, the different enzymes are produced at appropriate sites along the digestive system.

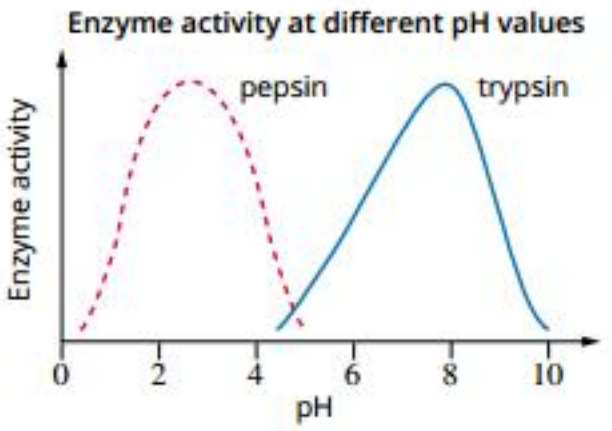


FIGURE 5.3.4 Pepsin and trypsin are both enzymes that digest proteins, but they have very different pH requirements. Pepsin is released in the stomach and is most active in its acidic environment. Trypsin is most active in the slightly alkaline small intestine.

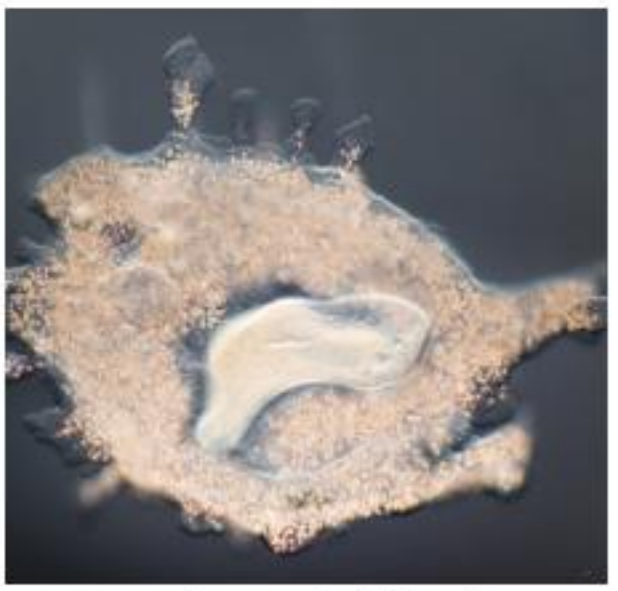


FIGURE 5.3.5 An example of intracellular digestion. This amoeba proteus is engulfing a *Paramecium caudatum* protozoan. Once ingested, the protozoan is contained inside a food vacuole into which lysosomes release digestive enzymes.

The importance of pH

Because enzymes are proteins, they are sensitive to changes in the pH of a solution (Figure 5.3.4). As you learnt in Chapter 3, altering the pH of the environment, changes the shape of protein molecules, which in turn alters their chemical properties. The change in shape alters the way that an enzyme binds with the molecule upon which it acts. Enzymes, therefore, have certain pH ranges over which they operate best. Different regions of the gut have different pH values that are most suitable for enzymes found in that region.

Extracellular digestion

Chemical digestion can be extracellular or intracellular. **Extracellular digestion** occurs when, for example, cells release enzymes into the **lumen** (central cavity) of the small intestine. There, enzymes split the food molecules and the resulting smaller molecules are absorbed. Extracellular digestion can also be seen in cells that excrete digestive enzymes like the pancreas. Sometimes digestive enzymes are located on the actual surface of cells. As the food is digested into smaller molecules, the molecules pass immediately into the cells. Mammals and most other animals rely on some form of extracellular digestion.

In contrast, many protozoans and invertebrate animals, such as mussels, sea jellies and free-living flatworms, use **intracellular digestion**. Their cells engulf small pieces of food into a membrane-bound food vacuole within the cell (Figure 5.3.5). Lysosomes fuse with the vacuole and release enzymes into it to digest the food. The resulting small molecules from the digestive process pass through the vacuole membrane and into the cytosol. Intracellular digestion is also seen in phagocytosis (see Module 2.4). Whilst this digestion is not to release food particles, it does, nevertheless, use enzymes to break down cells.

FEATURES OF EFFECTIVE DIGESTIVE SYSTEMS

In one sense, the digestive systems of all animals must be effective; otherwise, the animals would not exist. For each animal, their digestive system adequately provides for their needs. Large animals, including vertebrates, require higher levels of energy and nutrients for their normal activities. Because mammals are endothermic (they maintain a stable body temperature, usually higher than their environment), they require a lot of energy to maintain their body temperature. Therefore, they need digestive systems that can efficiently extract large amounts of energy and nutrients from food resources, and these systems are found in more active animals. Characteristics of these highly efficient digestive systems include:

- effective mechanisms for capture and preliminary handling of food
- appropriate physical breakdown of food
- a one-way gut with separation of tasks along its length
- efficient transport and storage of ingested food
- efficient sequential release of digestive enzymes
- an adequate surface area for maximal absorption of nutrients and water
- efficient egestion of unwanted materials.

DIGESTIVE SYSTEMS IN MAMMALS

All mammals need food and water, but different species (such as cows and dogs) have different food requirements, feeding behaviours and digestive systems. Cows are slow-moving and spend much of the day eating grass and chewing. In contrast, dogs are energetic and active, and may spend only 5–10 minutes each day gulping down food. Dogs and cows have many other differences that relate to their eating habits. Their teeth are very different, and cows have much larger and more complex intestines than dogs (Figure 5.3.6).

The feeding behaviour, teeth and digestive systems of cats and dogs are similar, but in guinea pigs and rabbits they are more like those of cows. One common factor is diet: cats and dogs eat meat, whereas cows, guinea pigs and rabbits eat plants.

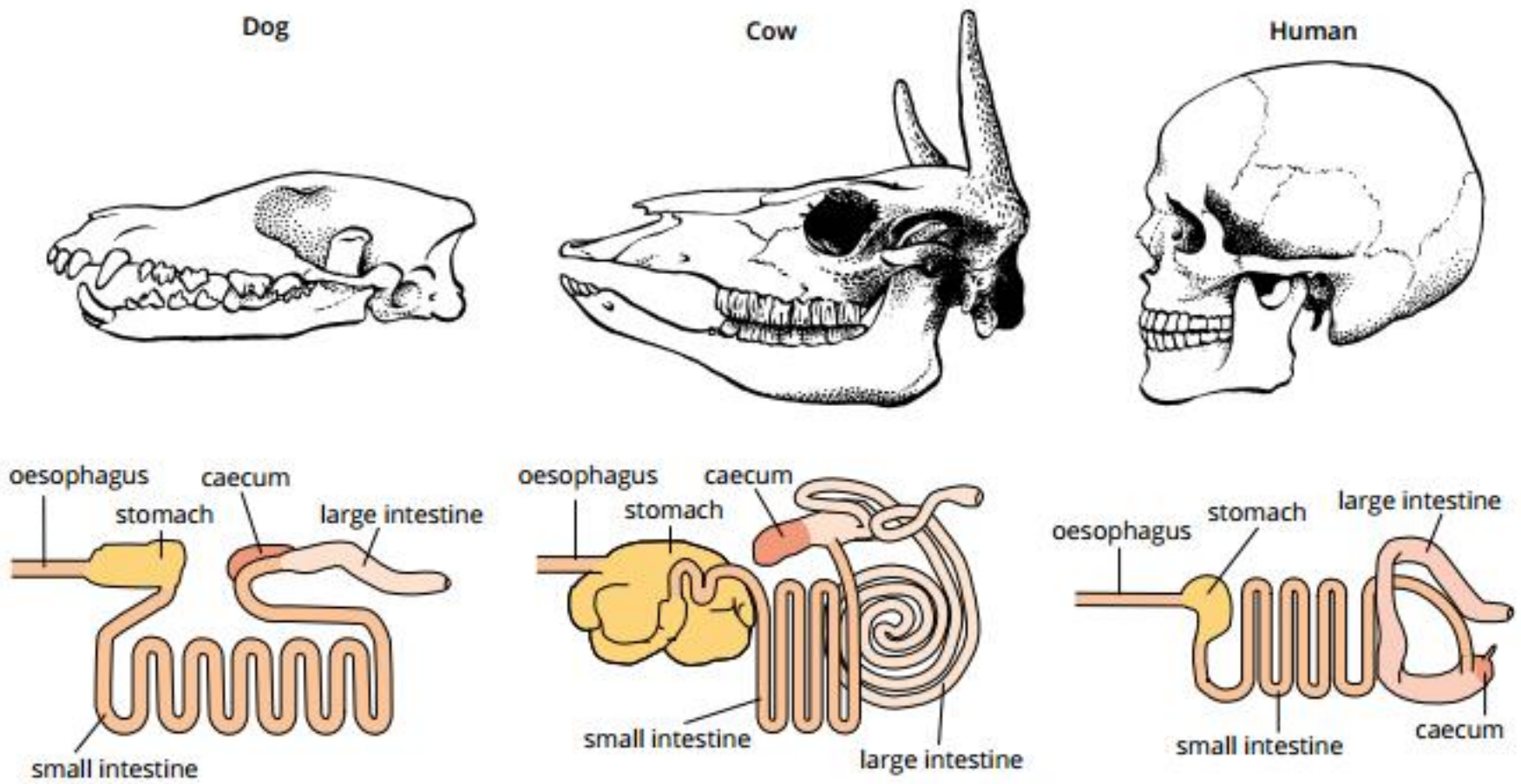


FIGURE 5.3.6 Skulls and digestive systems of the dog (a carnivore), the cow (a herbivore) and the human (an omnivore). Scientists studying fossil jaws, or even a few fossil teeth, can suggest the likely feeding behaviour and diet of the particular mammal species because teeth are modified in different species to suit the type of food that is eaten.

Humans are different again. Our teeth are unlike those of dogs or cows—we are not very good at chewing bones or grass! Our preferred foods include both meat and plant material, and we often cook our food first.

Humans spend 30–90 minutes each day eating, although the social aspects of eating may extend this time. The human digestive system is proportionally longer than that of a dog, but shorter than that of a cow.

Cows, dogs and humans are examples of animals with three different dietary patterns. Animals that eat only plants, such as cows, rabbits, kangaroos and koalas, are **herbivores** (Figure 5.3.7a). Herbivores typically spend much of the day eating. **Carnivores**, including dogs and cats, consume animals (Figure 5.3.7b). They spend much less time eating; sometimes animals in the wild, such as lions, may not eat for days between meals. Humans, on the other hand, are **omnivores** (from the Latin *omnivorus*, meaning 'eating everything'), because they can eat both plant and animal foods.

Animal matter has a much higher proportion of extractable energy per gram than plant matter. The carnivore gut produces all the enzymes needed for the complete digestion of meat. Digestion is quicker and more efficient. Digestive systems are shorter and simpler in carnivores than in herbivores.

The reason for the difference in feeding behaviour between herbivores and carnivores is clear. Plant material must be repeatedly ground by the teeth to expose as much surface area as possible for enzyme action and to release the contents from broken cells. As a food, plant material provides much less energy than meat and it takes a long time to extract that energy.

FIGURE 5.3.7 The skull of (a) a kangaroo—a herbivore, and (b) a Tasmanian devil—a carnivore

Human digestion

The primary function of the digestive system in mammals is to digest and absorb food. In other words, the digestive system breaks down food, making it simple enough to pass across cell membranes and be useful to cells.

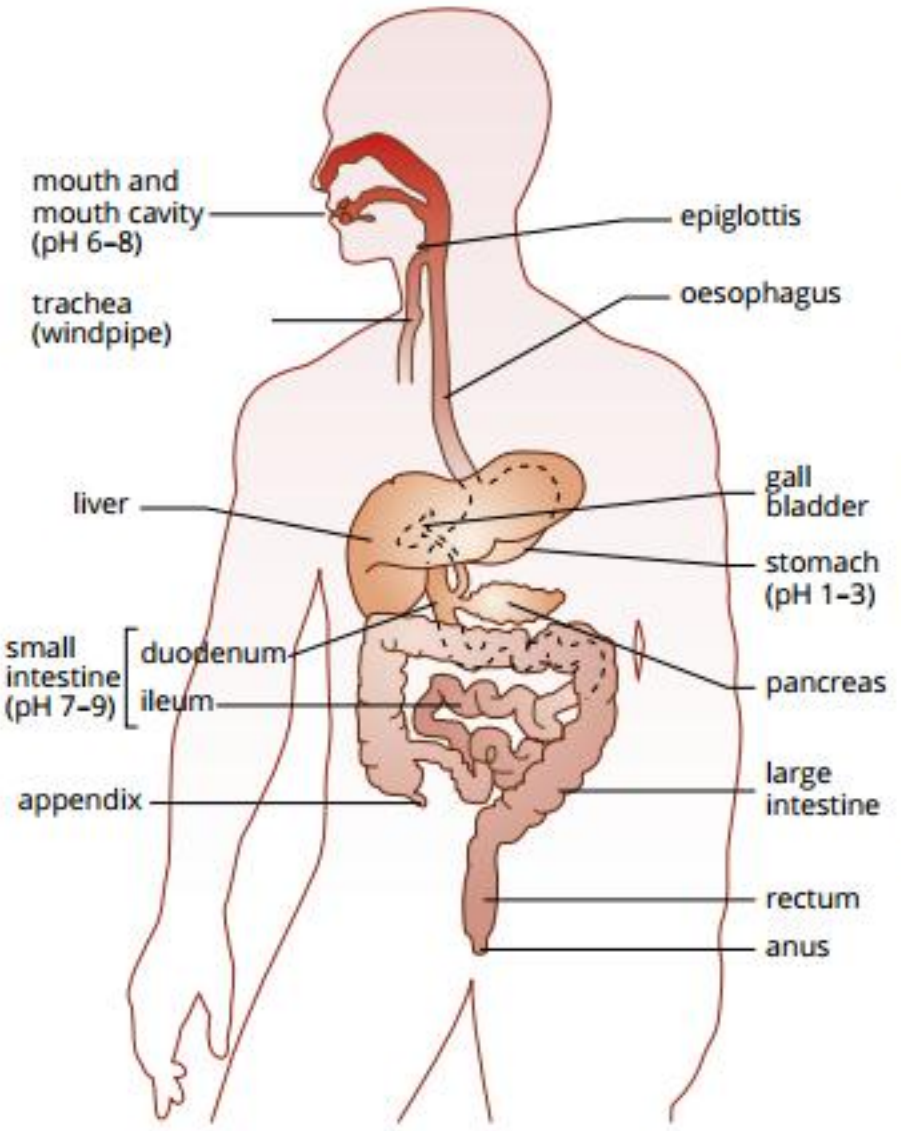


FIGURE 5.3.8 Components of the human digestive system

Before food passes into the digestive system of a mammal, it is physically broken into smaller pieces by teeth. Mucus is secreted to protect the lining of the gut and to lubricate food for easier passage. The food then moves along the gut past a series of digestive enzymes that sequentially break down the various compounds for absorption. Proteins are broken down to amino acids, fats and lipids to fatty acids and glycerol, and complex carbohydrates such as **starch** to simple sugars. Useful substances, such as water, are absorbed, leaving unwanted and undigested substances to be eliminated in the faeces.

The main regions of the human digestive system are the mouth and mouth (oral) cavity, oesophagus, stomach, small intestine, large intestine, rectum and anus. These components are shown in Figure 5.3.8. The salivary glands, pancreas and liver are digestive glands that develop as outgrowths (accessory organs) of the digestive system.

Key steps in the process of digestion in humans occur at the:

- mouth—teeth mechanically break food into small pieces. Saliva lubricates food and the enzyme, amylase, digests starch into maltose
- **epiglottis**—a flap, at the entrance to the larynx, prevents food from entering the trachea and respiratory system, directing it down the oesophagus. The epiglottis is also associated with the gag and cough reflex
- oesophagus—a tube down which food travels from the mouth to the stomach, aided by **peristalsis** (muscular contractions and relaxation)
- stomach—protein-digesting enzymes (proteases) and gastric juices (hydrochloric acid) are secreted to aid in food digestion. Peristalsis of the stomach muscles further mechanically breaks the food down and pushes digested food (chyme) into the small intestine
- liver—has important roles in regulating metabolism, toxin removal and processing nutrients. It stores excess glucose as glycogen (a polysaccharide or carbohydrate) for later conversion back to glucose when needed for energy. The liver is also the site of bile production, for the breakdown of fats
- gall bladder—stores and concentrates bile before releasing it into the small intestine
- pancreas—digestive enzymes are produced and activated when the food reaches the duodenum (first part of the small intestine). The pancreas also produces the hormones insulin and glucagon, which regulate sugar levels in the blood, and bicarbonate ions, which neutralises acidic chyme (acidic, partially digested food from the stomach)
- small intestine—the primary function is to absorb nutrients and minerals from food. Enzymes produced in the pancreas and the small intestine and bile from the liver and gall bladder further process food products to facilitate nutrient and water absorption. The small intestine's many blood vessels absorb the nutrients and waste products of digestion and deliver them to adjacent vessels of the circulatory system
- large intestine—water is absorbed with soluble compounds like vitamins and minerals; undigested food leaves the body as faeces.

Structure of the small intestine

The principal organ of absorption is the small intestine. 'Small' refers to the diameter of this part of the intestine. The small intestine is long and has a large surface area, making it well suited for absorption. The internal surface area is further increased by millions of tiny folds called villi, and also by the presence of many microvilli on the exposed surface of the epithelial cells lining the lumen (Figures 5.3.9 and 5.3.10).

Absorption in the small intestine

The epithelial lining in the small intestine is only one cell thick, allowing a rapid transfer of nutrients to the many blood and lymphatic vessels beneath the surface, which transport nutrients away to the body tissues. Nutrients pass through the lining of the small intestine by facilitated diffusion or active transport, along or against the concentration gradient.

Lipid-soluble molecules, which are the products of fat digestion (mainly fatty acids and glycerol), diffuse easily through the membranes of the epithelial cells along a concentration gradient. They then reassemble into fats before passing into the **lacteals**. Lacteals are capillaries of the lymphatic system near the intestine and have a milky appearance because of their high fat content after a fatty meal. Lipid-soluble vitamins (e.g. Vitamin A, D, E and K) also pass through the intestinal epithelium by passive diffusion.

Water-soluble molecules, including amino acids, simple sugars (monosaccharides such as glucose), and water-soluble vitamins (e.g. Vitamin C) and minerals pass through the membranes of the epithelial cells by active transport and facilitated diffusion. This can occur down or against a concentration gradient, ensuring that these essential nutrients are absorbed quickly.

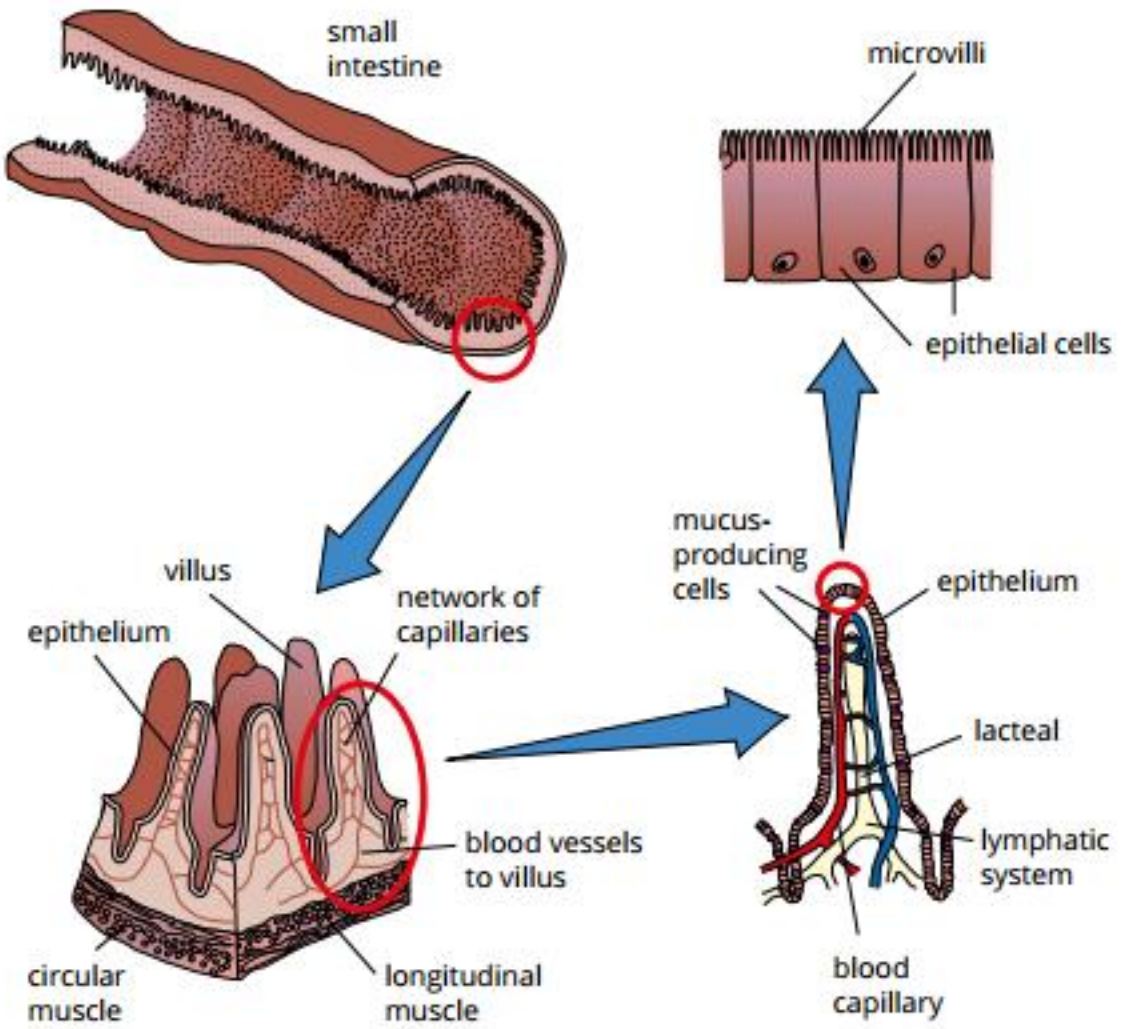


FIGURE 5.3.9 The internal surface of the human intestine, showing the villi and microvilli.

FIGURE 5.3.10 A cross-section of a villus (plural villi) from the small intestine. Villi are finger-like projections from the wall of the small intestine that increase the surface area for more efficient absorption of nutrients. Villi are covered in microvilli, which further increase the surface area for absorption.

Most of the water (90–95%) that enters the small intestine is also absorbed. This absorption is passive. Water diffuses across the lining of the intestine osmotically as the products of digestion are absorbed.

Blood leaving the intestine passes first into the liver through the hepatic portal vein, where absorbed nutrients can be removed and stored in the liver, before passing into the general venous circulation.

Herbivores utilise cellulose

Cellulose is the main component of plant cell walls, but its molecules are too large to be absorbed without digestion. Although many species of animals are herbivores, only a few can make the enzyme cellulase that is needed to digest cellulose. To get around this problem, herbivores have a symbiotic partnership, called mutualism, with bacteria that can produce cellulase. The bacteria live in the gut of the animal. They receive shelter and nutrients, and in return convert cellulose into simpler molecules that can be absorbed by the gut. The bacteria also supply important vitamins such as the B group and vitamin K.

The environment inside the gut is warm and wet but there is little or no oxygen, so the breakdown of cellulose must occur anaerobically by fermentation. Because of this, the part of the intestine in which the breakdown of cellulose occurs is sometimes called a fermentation chamber.

In herbivorous mammals, fermentation takes place in different parts of the intestine in different species, with varying degrees of efficiency. Generally, herbivorous mammals belong to either of two groups—hindgut or foregut fermenters.

Hindgut fermenters

In hindgut fermenters, fermentation occurs in the **caecum** (an enlarged pouch where the small and large intestines join), or the first part of the large intestine (the wombat in Figure 5.3.11), or both (the koala in Figure 5.3.11). Both of these are located after the small intestine, which is the region where most absorption takes place. This arrangement limits the advantage obtained from the symbiotic relationship, because the products of their digestion are not completely absorbed.

Horses are hindgut fermenters, and the relative inefficiency of their system can be seen by the fact that horse faeces contain large amounts of undigested plant material. Some hindgut fermenters, such as possums and rabbits, overcome this problem by producing two types of faeces. One of these comes directly from the caecum at night and is re-ingested so that it can go through the intestine again. This means that the vitamins and products of cellulose digestion from the bacteria are available for absorption from the small intestine.

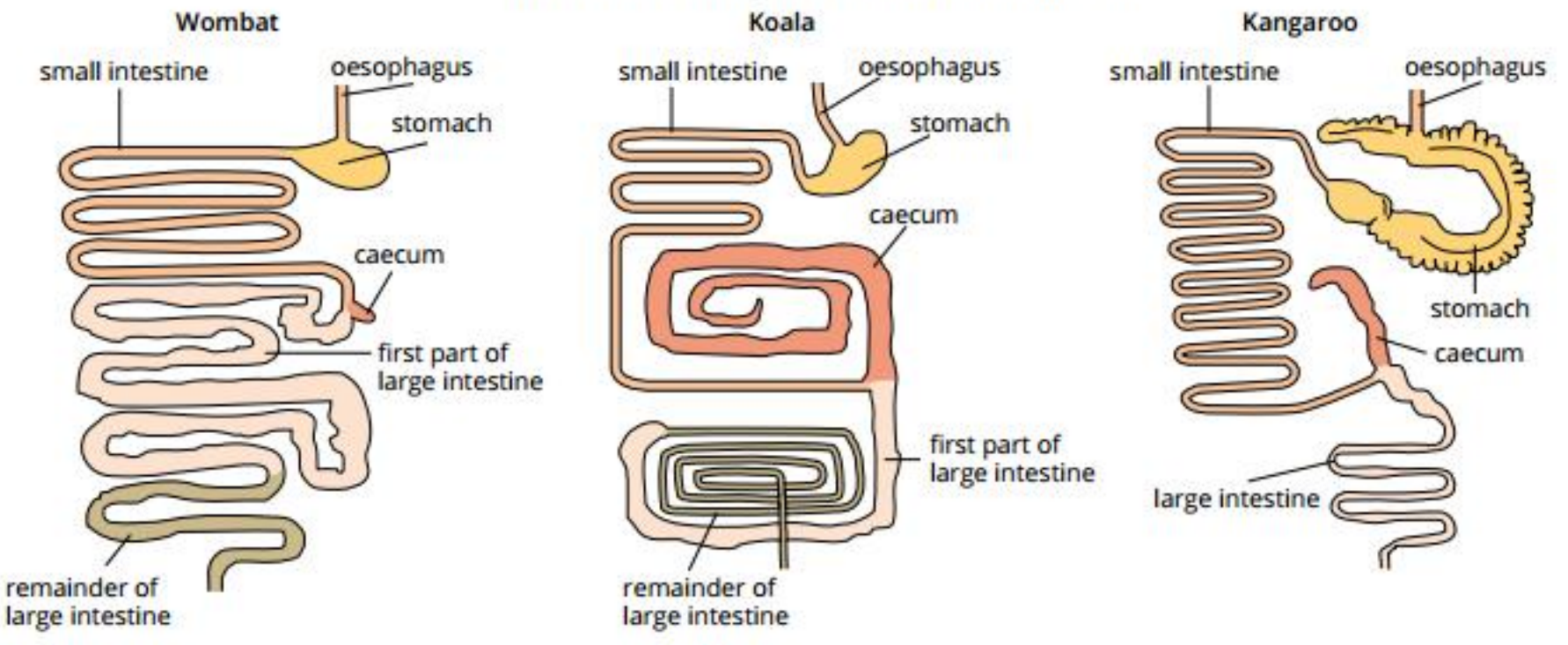


FIGURE 5.3.11 Wombats, koalas and kangaroos are herbivores and use symbiotic bacteria for the digestion of cellulose. Wombats and koalas are hindgut fermenters whereas kangaroos are foregut fermenters.

Foregut fermenters

In the foregut fermenters (such as the kangaroo in Figure 5.3.11), the fermentation chamber is located before the stomach. In ruminants such as cattle and sheep, it is called the **rumen**. Food can be regurgitated back into the mouth for further physical breakdown (rumination), then returned to the rumen for continued chemical breakdown by bacteria. This regurgitated food is called cud.

Foregut fermentation has the obvious advantage that the products of digestion by microorganisms are available for absorption along the entire length of the small intestine. Kangaroos and wallabies are the only marsupial foregut fermenters.

Ruminant digestion has some drawbacks. The complete digestion of plant material in the rumen by microorganisms can take a long time—hours or even days, with constant regurgitation and chewing of the cud. If the quality of food is very low (i.e. mostly cellulose and not much fresh, young plant growth), an animal may be starved of food that is digested enough for absorption, even though the animal has a very full rumen.

Food and energy storage in mammals

When food is not available, an animal's body draws on its own stores to meet its nutritional and energy needs. Energy storage is clearly essential for carnivores, which eat intermittently depending on the availability of prey.

Herbivores often have to travel considerable distances to find new and adequate supplies of the plants that they eat, when seasons change or if they have overgrazed an area. In winter, food generally becomes scarce for both herbivores and carnivores. In very cold climates, some mammals (usually small species) resort to hibernation to survive the winter (Figure 5.3.12). In each of these situations, the ability to store nutrients and energy reserves is essential for survival.

When needed, carbohydrate stores in the liver and muscles (glycogen) are used first and most easily, but their capacity is limited. Humans usually have enough **glycogen** stored to last for 12–48 hours of moderate activity. The capacity for storage in fat tissue (adipose tissue) is virtually unlimited (Figure 5.3.13). In a person of healthy weight, there is enough fat stored in tissues to allow normal energy consumption to continue for 3–7 weeks. As a last resort, the more limited protein stores of body tissues are used to provide energy after other stores are depleted.

Unlike carbohydrates and fats, amino acids cannot be stored in animal tissues, so animals need the full range of amino acids needed for building proteins in their diet. Proteins are assembled in cells by linking amino acids in a specific order. If the next amino acid required is not available, the synthesis of that protein molecule cannot continue until the required amino acid arrives.

This has consequences for strict vegetarians because, unlike meat, individual plants do not normally contain the full range of essential amino acids. However, by eating an appropriate combination of plant foods at the same meal, such as beans (which are a good source of the amino acids isoleucine and lysine, but deficient in tryptophan) and rice (which is deficient in isoleucine and lysine, but a good source of other essential amino acids), a balanced diet can be obtained. A meal of rice and beans together is as good a source of protein as eggs or meat.

FIGURE 5.3.12 The mountain pygmy-possum (*Burramys parvus*) is Australia's only hibernating marsupial. They accumulate fat stores before entering hibernation for 5–7 months every year.

FIGURE 5.3.13 A coloured scanning electron micrograph of a sample of fat tissue, showing fat cells (adipocytes) surrounded by fine strands of supportive connective tissue. Adipocytes are among the largest cells in the human body, each cell being 100–120 µm in diameter.

Energy reserves

In contrast to plants, animals have only a limited capacity to store carbohydrates. Carbohydrates are stored in animals as glycogen, which, like starch, is a large molecule made from glucose subunits. In humans, about 300 g of glucose is stored as glycogen in the liver and muscles. The remainder of our energy reserves is stored as fats.

Unlike plants, animals use fats rather than carbohydrates as their main form of energy reserves because:

- almost 25% more ATP is produced (per carbon atom) from fats than from carbohydrates
- fat is almost 50% lighter (per carbon atom) than carbohydrate
- stored carbohydrates attract and bind water molecules, increasing their weight by 200–500%; fats do not
- 1 gram of carbohydrate or protein provides up to 17 kJ of energy; 1 gram of fat provides 39 kJ of energy.

An average 70 kg male human stores about 11 kg of fat; the same amount of energy stored as carbohydrate could weigh more than 100 kg.

Some of the chemical processes that take place in living organisms use up energy, while other processes release energy. For energy balance, energy input (eating) must equal energy output (usage). If the amount of food eaten provides more energy than is used, the excess energy is stored as chemical energy (e.g. in fat or glycogen). If the energy content of food is less than required, the balance is made up from stored energy reserves or by breaking down body tissues.

REMOVING WASTES: THE EXCRETORY SYSTEM

As cells function using the nutrients provided, they produce substances that are no longer useful to them. The accumulation of these waste substances, such as carbon dioxide from respiration and **nitrogenous wastes** from protein breakdown, can prevent cells from functioning properly.

In mammals, the function of the kidneys is to excrete nitrogenous wastes. Excretion usually involves the loss of water and is, therefore, closely linked to water balance in terrestrial animals. This system works closely with the circulatory system, filtering waste products from the bloodstream and collecting them in urine.

For heterotrophs, it is sometimes inevitable that toxic substances are absorbed from the food they eat; these toxic substances must also be excreted. Excretion is the removal of substances that once formed part of the body of the organism. (This is different from egestion, which is the removal of undigested food from the gut in faeces.)

The internal environment of animals is extracellular fluid, and has a highly regulated composition. Salts form ions in solution. The concentrations of certain ions in cells are held within narrow limits. Some of these ions are also important for regulating the pH of body fluids, which must be at a suitable pH for enzymes and other molecules to function efficiently.

In animals, the removal of wastes and toxic substances, and the control of pH, ion concentrations and water balance, are carried out mainly by excretory organs. These processes vary with the activity of the animal and the external conditions.

The nature of wastes

During normal activity, animal cells break down and replace carbohydrates, lipids and proteins, producing waste products that usually cannot be used by the body.

Carbon dioxide

When carbohydrates or lipids are broken down during cellular respiration to release energy, carbon dioxide and water are produced (see Module 3.5). These are released into the surrounding environment across respiratory membranes, which in mammals are in the lungs. Water produced during cellular respiration is incorporated into body fluids, and excess water is expelled from the body.

Nitrogenous wastes

Protein consists of amino acids, which contain nitrogen. When proteins are broken down, the nitrogenous parts are split off and the remainder of the molecule is converted into carbohydrates or lipids, which can be used for energy. The remaining nitrogenous wastes must be removed from the cell, because they can become toxic.

The first nitrogenous waste to be formed from the breakdown of protein is **ammonia**. Ammonia is highly soluble in water, and many aquatic organisms take advantage of this solubility to effectively remove nitrogenous wastes as ammonia from their body, often through the gills. One problem with ammonia is its toxicity to cells, even at low concentrations; ammonia cannot be stored in the body for any length of time.

Ammonia can be converted into two less toxic forms, **urea** and **uric acid** (Figure 5.3.14), but this process requires energy. Neither urea nor uric acid is of any further use to most animals' however, because urea and uric acid are less toxic than ammonia, they can be stored in the body before being excreted. Mammals excrete nitrogenous waste in the form of urea, which is in urine. Reptiles and birds excrete nitrogenous waste in the form of uric acid. The white component of bird droppings contains uric acid crystals.

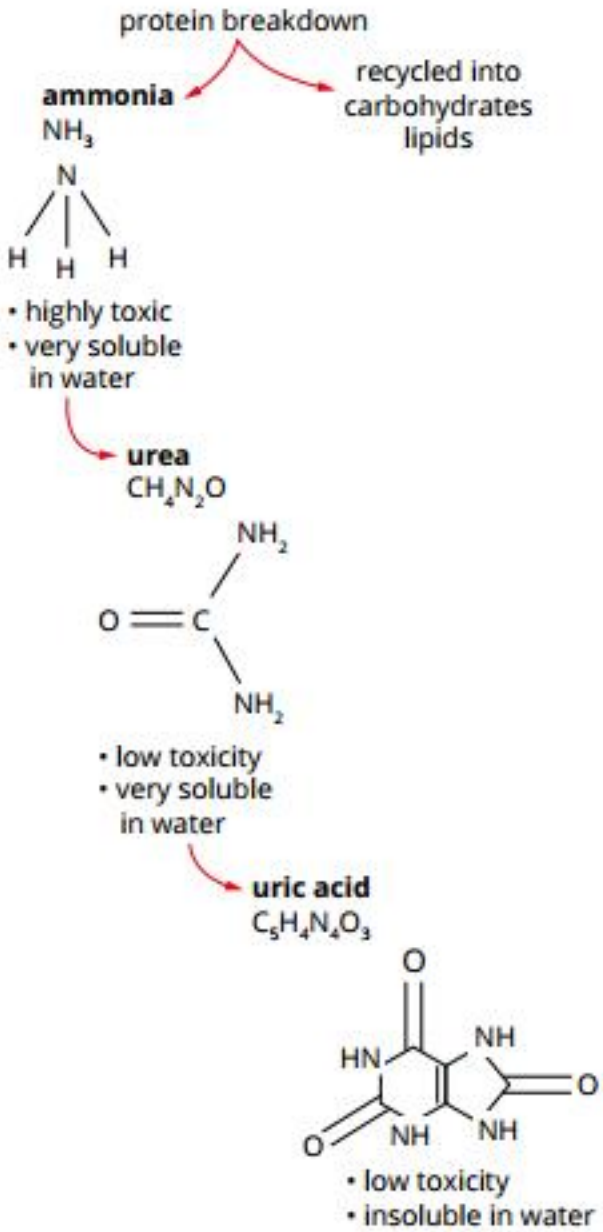


FIGURE 5.3.14 Three important nitrogenous wastes produced from the breakdown of proteins in animals. Mammals excrete their nitrogenous wastes in the form of urea.

Excretory mechanisms in mammals

Kidneys

The kidneys of all vertebrates, from fishes to mammals, function by filtering blood, then reabsorbing useful substances and secreting unwanted ones. Blood is filtered through blood vessel walls to form a primary filtrate that has the same composition as plasma, except for the large proteins that have been filtered out. Most of the useful substances in the primary filtrate are reabsorbed as it passes through the kidney tubule. Some unwanted substances (such as urea, Na^+, K^+, HCO^{3-} depending on the body's needs) may be secreted into the fluid in the tubule before it passes out of the kidney to the **bladder**. These processes regulate the concentration of different salts in the blood, including those salts that are responsible for maintaining the pH of body fluids within closely controlled limits.

Mammals conserve water by producing urine that is more concentrated than body fluids. The ability to produce concentrated urine is related in some mammals to the degree of water stress experienced in their normal environments. Desert-adapted mammals, such as the bilby in Figure 5.3.15, are able to excrete highly concentrated urine.

The liver prepares wastes

The liver performs many different functions and has a central role in the maintenance of a stable internal environment. In addition, it is responsible for preparing various substances for excretion. It detoxifies a variety of harmful chemicals such as alcohol and some drugs. It is also responsible for breaking down amino acids to release ammonia, which it then converts largely into urea. The waste products from these processes travel in the bloodstream to the kidneys for excretion.

The liver also destroys worn-out red blood cells, producing bile pigments from the breakdown of haemoglobin. Bile pigments, along with bile salts, which emulsify fats as part of digestion, are stored in the gall bladder before they are released into the lumen of the small intestine. Bile pigments are one of the few substances excreted into the gut.

FIGURE 5.3.15 Desert-dwelling animals such as the bilby (*Macrotis lagotis*) can produce highly concentrated urine, which minimises water loss.

THE MAMMALIAN KIDNEY

Mammals have two kidneys at the back of the abdominal cavity. There is always a high blood flow to the kidneys because they are important in maintaining the stability of the internal environment. Although kidneys are only about 1% of body tissue, they filter approximately 20–25%' of the body's blood every minute.

Blood enters the kidney from the aorta through the renal artery, and leaves through the renal vein. Blood vessels branch throughout the kidney in a complex fashion (Figure 5.3.16). Urine, formed in the kidneys, drains via the **ureters** into the bladder, for storage, until an appropriate time for release through the **urethra**.

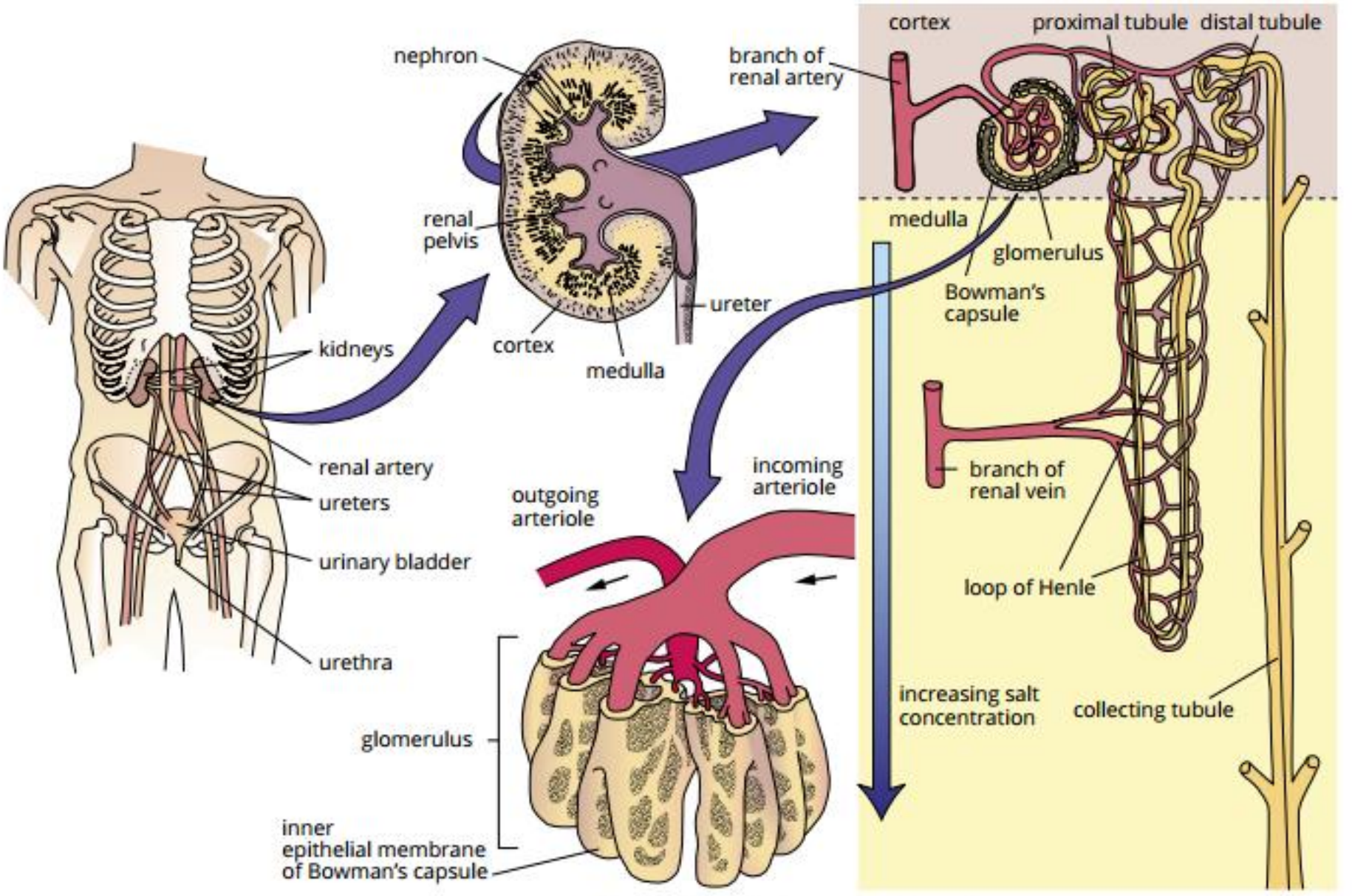


FIGURE 5.3.16 The structure of the human excretory system

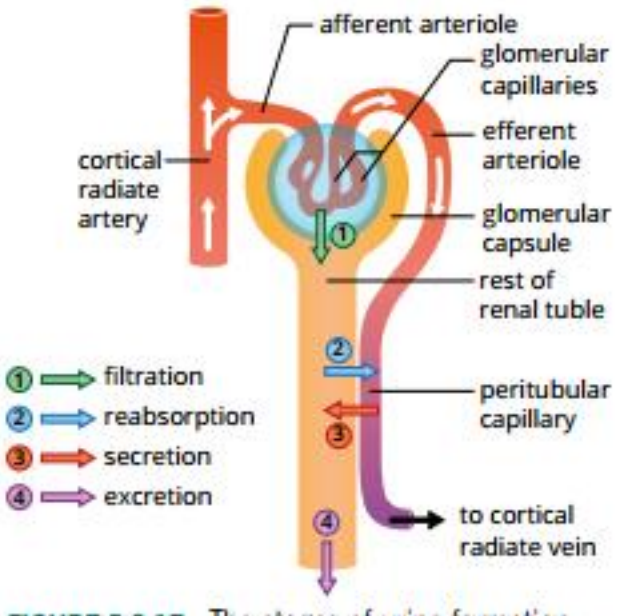


FIGURE 5.3.17 The stages of urine formation involve filtration, reabsorption, and secretion. Urine is stored in the bladder before it is finally excreted.

The functions of the mammalian kidney are carried out by **nephrons**, which are the functional units of the kidney. There are approximately one million nephrons in a human kidney, and their combined function carries out the work of the kidney. A nephron is composed of a **Bowman's capsule** surrounding a **glomerulus**, and a tubular region consisting of the proximal convoluted tubule, **loop of Henle** and distal convoluted tubule that leads into a collecting tubule (Figure 5.3.16). The formation of urine involves passive filtration, selective **reabsorption** and **secretion**, and the passive removal of water.

The nephron is very closely associated with blood vessels, particularly the glomerulus, which is a clump of looping capillaries embedded in the Bowman's capsule, and networks of capillaries wrapped around the remainder of the tubule.

There are two distinct regions in the kidney: the outer cortex and the inner medulla. Glomeruli are located in the cortex (Figure 5.3.17).

Filtration

Filtration occurs across the glomerulus into the Bowman's capsule, as shown in Figure 5.3.18. The high pressure of blood in the glomerular blood vessels forces fluid through the walls of glomerular capillaries and into the Bowman's capsule.

Only small molecules and water can pass through the wall membranes; blood cells and large blood proteins remain behind in the glomerular capillaries. This primary filtrate has the same composition as blood plasma, without large proteins.

If red blood cells or large proteins are found in urine, this indicates that the normal filtration mechanism has broken down and blood is leaking from the glomerulus into the Bowman's capsule. This may occur as a result of damaged glomerular blood vessels, or very high blood pressure.

FIGURE 5.3.18 A coloured scanning electron micrograph (SEM) of a glomerulus—a tight knot of blood capillaries (yellow) inside the Bowman's capsule (pink)

Reabsorption

Approximately 99% of the primary filtrate—including salts, glucose, amino acids and water, but only half or less of the urea—undergoes reabsorption along the length of the nephron (Figure 5.3.17). Virtually all amino acids and glucose are reabsorbed in the proximal convoluted tubules by active transport against a concentration gradient. The presence of glucose or amino acids in urine, therefore, indicates a possible kidney malfunction. Specific salts, particularly sodium chloride, are also actively reabsorbed. These active processes consume a lot of energy.

Water is reabsorbed from the urine passively, along an osmotic gradient. The mechanism by which the kidney is able to produce concentrated urine involves the loop of Henle (Figure 5.3.19). A large amount of sodium chloride pumped out of the loop of Henle is retained in the medullary region of the kidney, producing a very high salt concentration. The osmotic concentration within the kidney therefore increases considerably from the outer cortex to the medulla. When the urine finally passes down the collecting tubules towards the ureter, it passes through this region of high salt concentration. Because the collecting tubule is permeable to water, but not to salt, water passes from the collecting tubule back into the kidney and into blood vessels. As a result, the urine becomes concentrated. **Antidiuretic hormone (ADH)** from the pituitary gland increases the permeability of the collecting tubule to water, increasing reabsorption of water back into circulation and causing urine to become concentrated. This will be explored further in Chapter 6.

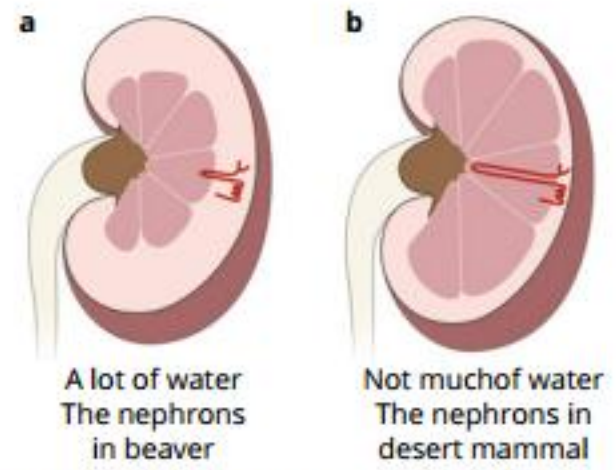


FIGURE 5.3.19 The loop of Henle in (a) an animal (e.g. beaver) that has access to a lot of water and (b) an animal (e.g. desert mammal—camel) that has access to little water. The loop of Henle is much longer in animals that live in desert environments, allowing them to conserve water by producing hyper-concentrated urine.

The descending limb of the loop of Henle is permeable to water thus more water is reabsorbed into the blood instead of staying in the filtrate. The longer the loop of Henle, the greater the reabsorption of water. This is important for desert animals who have little opportunity to find water in their environment. Animals in dry environments have very long loops of Henle to ensure they conserve the maximum amount of water in their bodies rather than lose it in their urine. This can be seen in Figure 5.3.19 where the beaver has a moderate length loop of Henle, but the desert mammal has a very long loop of Henle.

Secretion

Secretion is the active removal (excretion) of particular substances by the cells of the tubule wall (Figure 5.3.17). Ammonium, potassium and hydrogen ions are actively secreted into the distal convoluted parts of the tubules. Various dyes and drugs such as penicillin and aspirin are also eliminated by tubular secretion. These substances are added to the filtrate as it passes through the nephron. The filtrate is then carried from the bladder and excreted from the body as urine by the ureter.

5.3 Review

SUMMARY

- Mammals are heterotrophs. They must consume other organisms or their products to obtain organic molecules.
- The purpose of digestion is to rapidly break down organic food into molecules small enough to be able to pass through membranes and into cells.
- Chemical digestion involves breaking apart complex molecules into simple molecules by the action of enzymes (amylase, protease and lipase).
- Physical breakdown of large food into smaller pieces increases the surface area available for enzyme action and therefore increases the efficiency of digestion.
- When food is not available, an animal's body draws on its own stores to meet its nutritional and energy needs.
- Mammals can store excess carbohydrates and fats, but not amino acids. Lipids store more energy by weight than carbohydrates.
- For energy balance, energy input (eating) must equal energy output (usage).
- Excretion is the removal of substances that once formed part of the body of the organism.
- In animals, removal of waste and toxic substances, and control of pH, ion concentrations and water balance, are carried out largely by excretory organs, such as the kidney.
- Proteins are broken down and converted into carbohydrates or lipids, which can be used for energy, and nitrogenous wastes, which must be removed from the cell, because they can become toxic.
- The nephron is the functional unit of the mammalian kidney.
- The three main stages of urine formation are filtration, reabsorption and secretion.
- A nephron consists of a Bowman's capsule (surrounding a glomerulus) leading into a tubular region (proximal convoluted tubule, loop of Henle and distal convoluted tubule) and then into the collecting tubule.

KEY QUESTIONS

Describe

1 Define 'digestion'.

2 Recall three characteristics of an efficient digestive system.

3 Name the two main types of wastes produced by mammals.

4 Identify the key elements of cells in the small intestines which allows for efficient exchange of digestive material.

5 Describe how herbivores are able to breakdown the cellulose material in their diet.

6 Describe two ways, with examples, that maximise surface area in mammalian digestive systems.

Apply

7 Create a table to outline where the main digestion of biomacromolecules occurs within the human digestive system.

8 Identify which sections of the nephron sit in the cortex and which sit in the medulla.

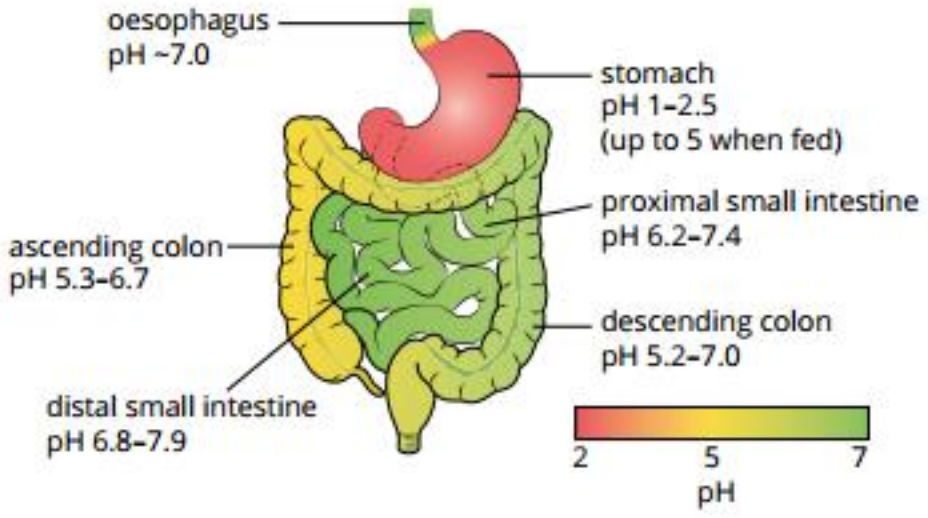
9 Explain the main differences between chemical and physical digestion.

10 Explain why it is important that the permeability to water of the collecting tubule of the mammalian kidney can be regulated.

Analyse

11 Organise the following to show the path of filtrate through a renal tubule: ascending limb of loop of Henle, descending limb of loop of Henle, distal convoluted tubule, proximal convoluted tubule.

12 The following diagram shows the pH of the human digestive tract. Pepsin is an enzyme that is released into the stomach to break down protein. Deduce what happens to pepsin once it leaves the stomach.

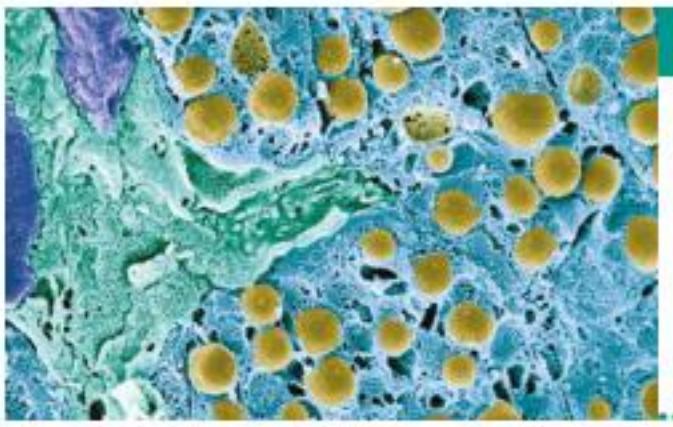


13 Compare the excretory functions of the mammalian kidney and liver.

14 Dialysis is used by patients with kidney failure. During dialysis, small solute molecules pass through a partially permeable membrane, but larger proteins and cells do not. Identify which state of normal kidney function dialysis replicates.

15 The pancreas plays some important roles in digestion. Explore these roles and infer what might happen to a person if their pancreas was damaged.

5.4 Gas exchange and transportation in vascular plants



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain how vascular plants exchange gases with their environment
- explain the role of stomata in gas exchange and water regulation in plants
- compare how xylem vessels transport water and nutrients from the roots to the leaves whilst phloem vessels transport sugars produced during photosynthesis from the leaves to the roots
- identify factors that affect the transpiration stream within plants.

Plants carry out two energy-transforming processes. Cellular respiration occurs throughout all of the cells in **vascular plants**, requiring oxygen and producing carbon dioxide. However, photosynthesis occurs primarily in cells in the leaves, where chloroplasts in the cells convert water and carbon dioxide, in the presence of sunlight, into glucose, water and oxygen.

Vascular plants include ferns, cycads, conifers and flowering plants and usually grow in terrestrial environments. Like complex animals, vascular plants have specialised tissues and organs to exchange gases and move substances, such as nutrients and wastes, around the organism. Vascular plants are characterised by the presence of **vascular tissue,** which is tissue that is specialised for transporting fluids. Two of the major organs in plants are leaves and roots. In vascular plants, vascular tissue is found within both of these organs.

In vascular plants, gas exchange occurs in the leaves, stems and roots. Unlike animals, plants do not have a specific mechanism for gas exchange, but can absorb or release gas directly from their environment. Transport within vascular plants occurs inside closed vessels organised into **vascular bundles** that move water, mineral ions and sugars around the plant. They consist of:

- **xylem vessel**s, which transport water and nutrients to the leaf from the roots
- **phloem** vessels, which transport sugars produced during photosynthesis from the leaves to the roots
- a sheath of **lignin**, which strengthens and supports the tissue.

In this module, you will learn about the tissues and organs in vascular plants that exchange gases, and transport water and mineral ions.

GAS EXCHANGE IN VASCULAR PLANTS

Most plants do not have specialised organs for gas exchange. Simple plants, such as mosses, have leaves that are small and extremely thin, only one cell thick, so each cell is in direct contact with the surrounding environment. Gases such as oxygen and carbon dioxide can easily diffuse directly between the air and the contents of each cell. In vascular plants (plants with conducting tissue), the exchange of oxygen and carbon dioxide in the leaves, stems and roots occurs by diffusion through special openings in the epidermis called **stomata** (sing. stoma).

The rate of movement of gases between air spaces and the atmosphere is regulated by the stomata, the main route through which gas exchange occurs. When the stomata are closed, the exchange of oxygen, carbon dioxide and water vapour between the plant and its environment virtually stops. Only small quantities of gases are able to pass directly through the epidermis and the overlying cuticle (the outer waxy layer).

Plant cells are loosely packed, allowing rapid diffusion of gases through intercellular spaces, which are filled with air (Figure 5.4.1). During gas exchange, oxygen and carbon dioxide diffuse from these air spaces through the water film covering the cells and into the cells along concentration gradients. Diffusion also occurs in the reverse direction.

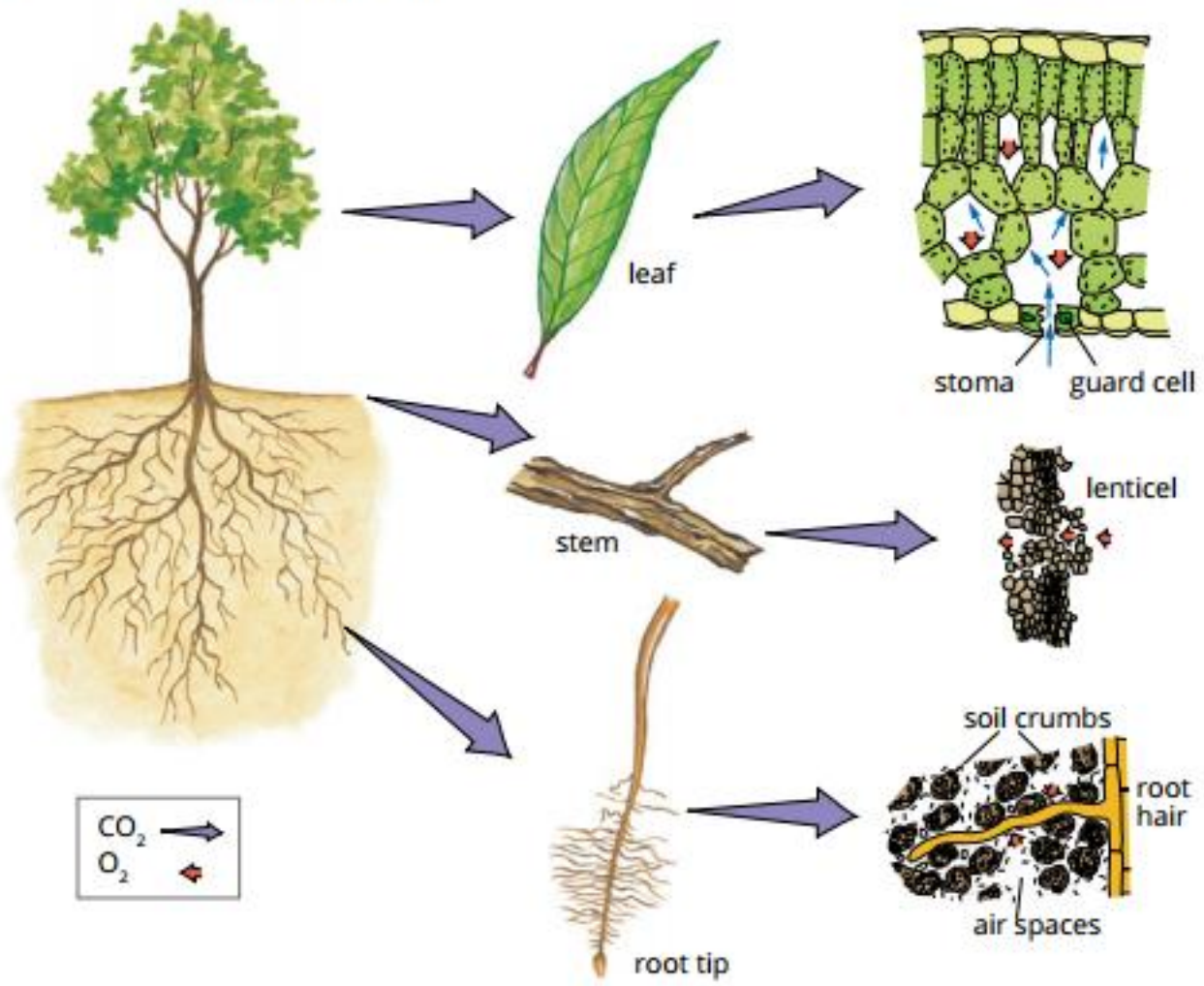


FIGURE 5.4.1 Routes of gas exchange with cells of leaves, stems and roots

Because diffusion distances are not great, and cells are organised in thin layers near the surface (even in large plants), gases do not generally have to be transported from one part of a plant to another.

Stomata

Stomata are tiny pores in the epidermis, bordered by two highly specialised epidermal cells called **guard cells** (Figure 5.4.2). Unlike other epidermal cells, guard cells contain chloroplasts. Stomata can occur on any part of a plant except the roots, but in most species, they are most abundant on the surface of the leaves.

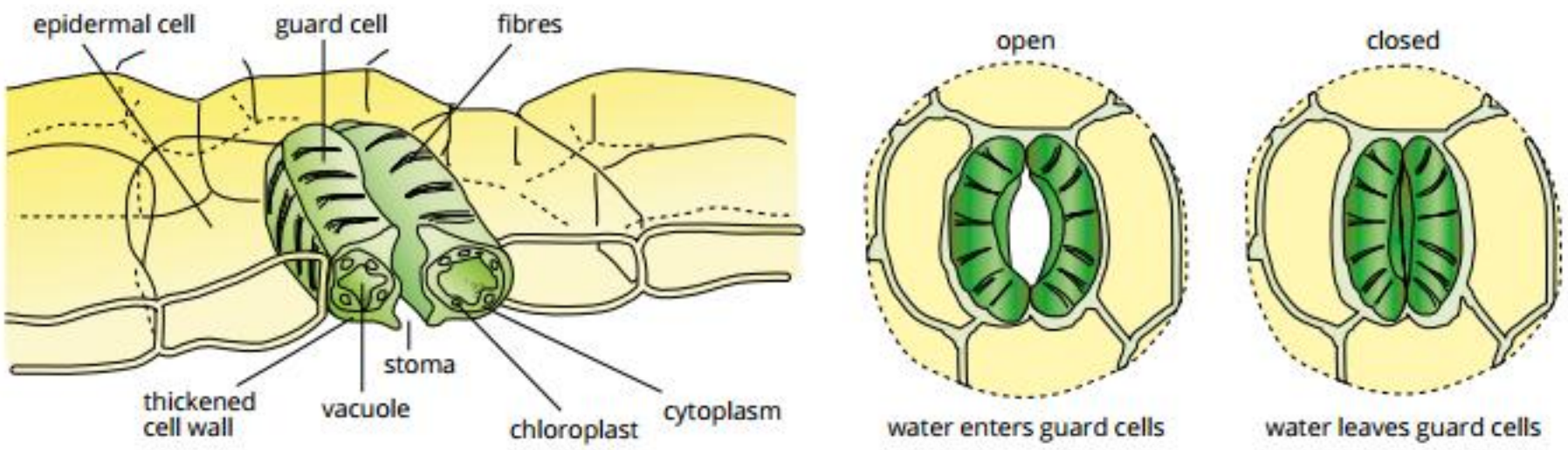


FIGURE 5.4.2 Gas exchange in leaves occurs through stomata. When water enters guard cells, they expand, opening the stoma. Guard cells expand lengthwise because they have a thickened inner cell wall containing cellulose fibres that prevent the cells expanding in width.

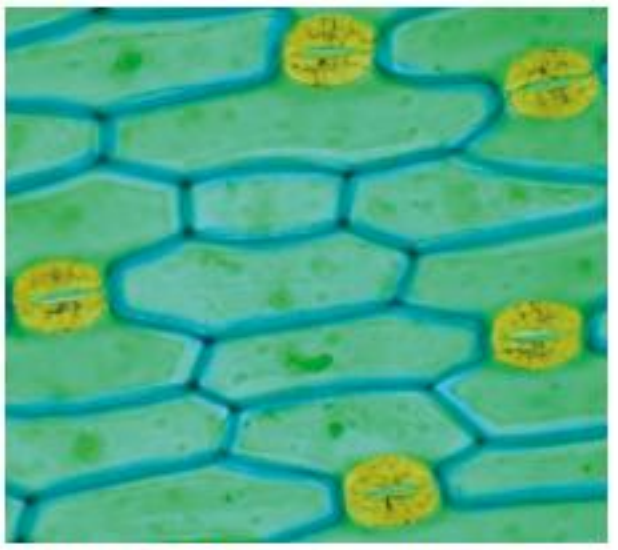


FIGURE 5.4.3 A light micrograph of stomata on the surface of a plant. Guard cells (yellow) either side of the opening regulate the exchange of gases and water vapour into and out of the plant.

FIGURE 5.4.4 A cross-section through the surface of a woody stem showing a loosely packed lenticel on the left bordered by layers of waterproof cork cells.

The number and size of stomata on a leaf vary according to the plant species and the environmental conditions under which it has grown.

In a typical plant, most stomata are on the underside of the leaves, away from the drying effect of the sun's rays. In contrast, stomata in floating aquatic plants, such as water lilies, are confined to the upper epidermis. In plants such as eucalypts, which are adapted to dry conditions, stomata are often in sunken pits in the surface of the leaves. This reduces direct flow of air across them and so reduces water loss.

Controlling guard cells

Guard cells (Figures 5.4.2 and 5.4.3) have the following structural features relating to their function:

- They are joined at their ends in pairs.
- Their cell walls are thicker on the side adjacent to the stoma.
- Bands of inelastic fibres run around each cell wall.

When water passes into the guard cells, their internal fluid pressure, or **turgor**, increases. This causes them to expand in the only direction possible: lengthways. The guard cells buckle and open the stoma. Conditions favouring the opening of stomata are abundant water, light and low internal carbon dioxide concentrations.

Terrestrial plants, like terrestrial animals, must reduce loss of water by evaporation. The moist surfaces that they use for gas exchange are the major site of water loss. Therefore, the stomata act to balance the plant's need to obtain carbon dioxide for photosynthesis against the dangers of drying out due to the loss of water from the leaves.

During daylight, when plants are photosynthesising, large volumes of carbon dioxide and oxygen are exchanged with the environment through open stomata. At night, when photosynthesis is not occurring, stomata are usually closed. Stomata also close during the day if it is very hot and dry. This prevents excessive water loss, but also drastically reduces the rate of photosynthesis.

Thus, conditions that usually favour the opening of stomata are abundant water, bright light and low internal carbon dioxide concentrations.

Stems and roots

In the epidermis of green stems, as in the leaf epidermis, there are stomata through which gas exchange takes place. In woody stems and mature roots, the epidermis is replaced by a layer of cork cells that are waterproof and airproof. Air passes freely through groups of these loosely packed cells to the cells beneath. Each group of loosely packed cells is called a **lenticel** (Figure 5.4.4).

Roots exchange gases with the air in spaces in the soil. Oxygen readily diffuses into the film of moisture surrounding **root hairs**, and then into the roots themselves. When soil is waterlogged due to excessive rain or poor drainage, the spaces in the soil are filled with water instead of air. Because the amount of oxygen dissolved in water is so much less than the amount of oxygen in air, the roots may not be able to get enough oxygen for their needs. The root cells may die, killing the plant. Many indoor plants are 'killed with kindness' by over-watering.

Aquatic plants

In aquatic plants, roots are often continuously submerged. In some of these species, oxygen diffuses from aerial parts of the plant into the submerged organs. Some aquatic plants have adaptations that help in gas exchange. Pondweeds and water hyacinths float on water because the leaf stalks are swollen with large, air-filled intercellular spaces (Figure 5.4.5a). Mangroves, which live in salty, waterlogged mudflats, have specialised roots called pneumatophores that assist in gas exchange (Figure 5.4.5b). Pneumatophores also contain large intercellular spaces.

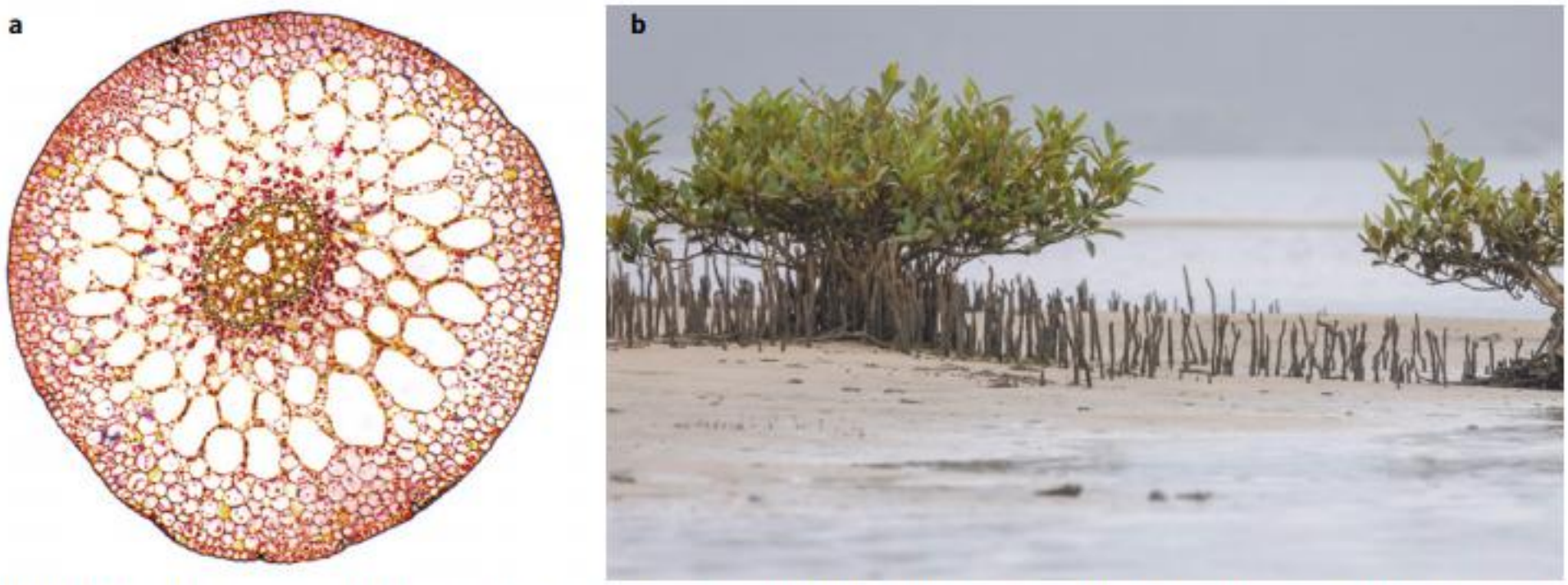


FIGURE 5.4.5 (a) The pondweed (*Potamogeton*) has long stems and broad floating leaves. This cross-section of its stem shows many air-filled intercellular spaces. (b) Mangroves send up specialised roots (pneumatophores) to help with gas exchange.

TRANSPORTATION IN VASCULAR PLANTS

Like all plants, vascular plants are autotrophs or producers, manufacturing their food from light energy by photosynthesis. For photosynthesis, plants need water, carbon dioxide and sunlight for energy. Water is absorbed through the roots, and carbon dioxide is absorbed via the leaves. Photosynthesis occurs in the leaves and produces the sugars that are needed by all active cells of the plant. Transport of these substances to the locations where they are needed is made possible by the presence of vascular tissue (Figure 5.4.6).

In large vascular plants, the leaves can be a long way from where water is absorbed via the roots, and the active cells in the roots requiring sugars for the energy requirements of nutrient uptake can be a long way from where photosynthesis occurs in the leaves. In tall trees such as the Californian redwoods pictured in Figure 5.4.7, water and nutrients need to be transported more than 100 metres from the roots to the upper branches! Their root system is relatively shallow, but water still needs to travel a great distance from where it is absorbed via the roots and root hairs to the leaves, where it is used during photosynthesis.

FIGURE 5.4.6 The veins in a leaf are vascular tissue that is specialised for transporting water and organic solutes throughout the plant. Each vein is made up of a vascular bundle of xylem, phloem and a lignin sheath.

FIGURE 5.4.7 The tallest trees in the world are the giant sequoias (*Sequoiadendron giganteum*) of California. These trees can reach a height of more than 115 metres above the ground.

Vascular tissue transports:

- water and mineral ions obtained from the soil by the roots throughout the plant
- sugars made in the leaves to other parts of the plant.

In vascular plants, there are two types of vascular tissue.

- **Xylem** transports water and inorganic nutrients (mineral ions) absorbed from the soil up the plant.
- **Phloem** transports dissolved sugars, which are produced in the leaves by photosynthesis, throughout the plant. Other organic substances, such as amino acids are also transported in the phloem.

Xylem and phloem contain continuous, closed tubular pathways through roots, stems and leaves (Figure 5.4.8). Fluids flow through these tubules to all parts of the plant. All cells are close to vascular tissue.

The arrangement of xylem and phloem tissues in roots, stems and leaves is distinctive. Roots have a central core of xylem in a star or cross shape, with phloem between the arms of the xylem. In stems and leaves the xylem and phloem are grouped into vascular bundles. Figure 5.4.9 illustrates some different ways that xylem and phloem could be arranged in plants. Vascular bundles extend into the leaves.

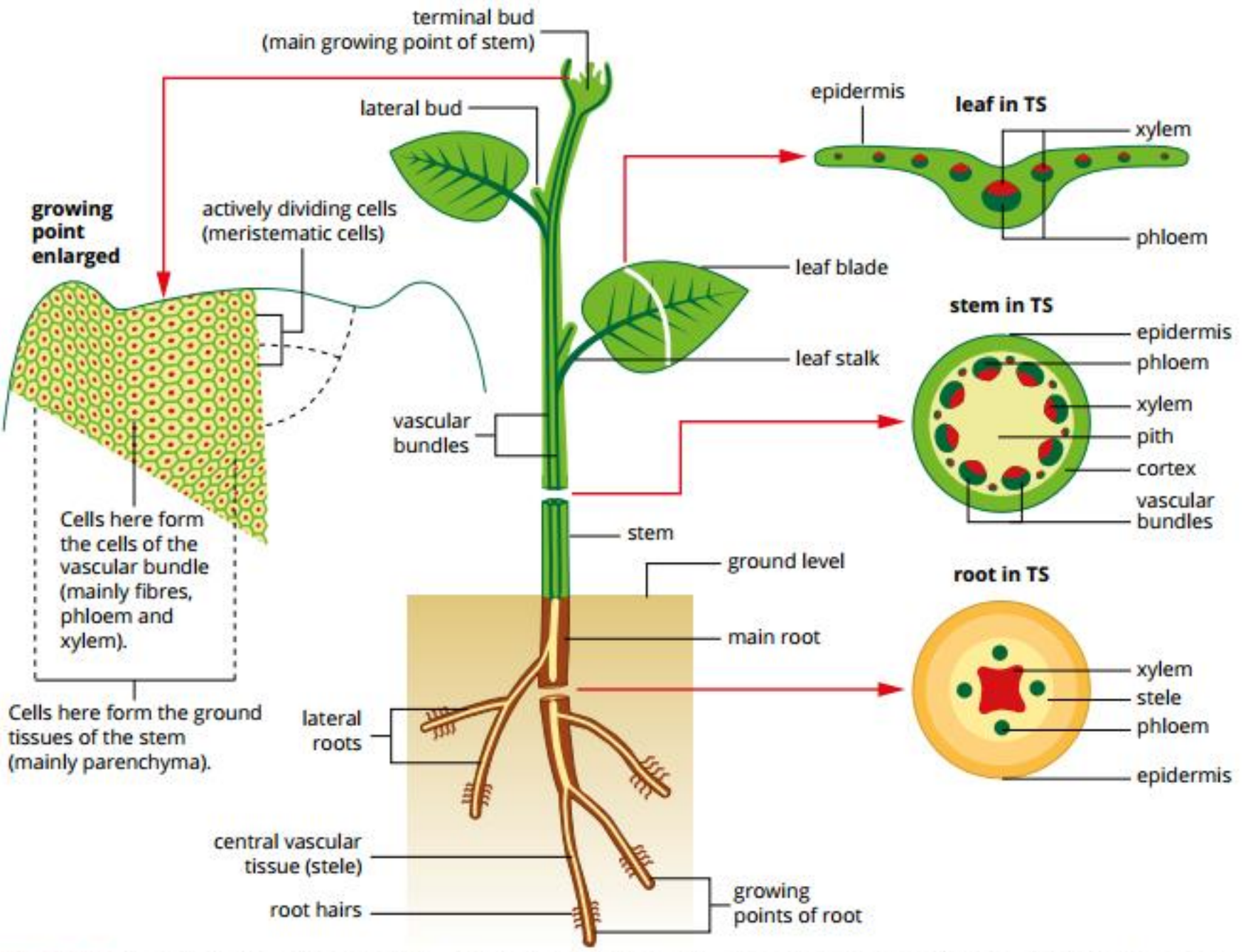


FIGURE 5.4.8 The main function of the transport system in plants is to transport water and organic solutes from the roots to the leaves and to transport food manufactured in the leaves (sucrose and amino acids) to the other plant tissues. The transport system tissues, the xylem and phloem, are continuous, tubular pathways through the roots, stems and leaves. (TS means transverse section.)

Xylem

Xylem is the vascular tissue that transports water and mineral ions obtained from the soil throughout the plant. It is composed mainly of xylem vessels and **tracheids**.

A mature xylem vessel is a long, water-filled tube consisting of elongated cells joined end to end (Figure 5.4.10). As these cells mature, the cell wall is strengthened with lignin, becoming stronger and more rigid, and the cytoplasm and nucleus disintegrate. Mature xylem vessels have:

- cylindrical skeletons of dead cells joined end to end to form continuous tubes
- perforated or complete openings at each end, like a straw, so that fluid can flow directly through them
- pits (non-thickened areas) and perforations in the sidewalls that allow sideways movement of substances between neighbouring vessels in the vascular bundle
- no nucleus or cytoplasm.

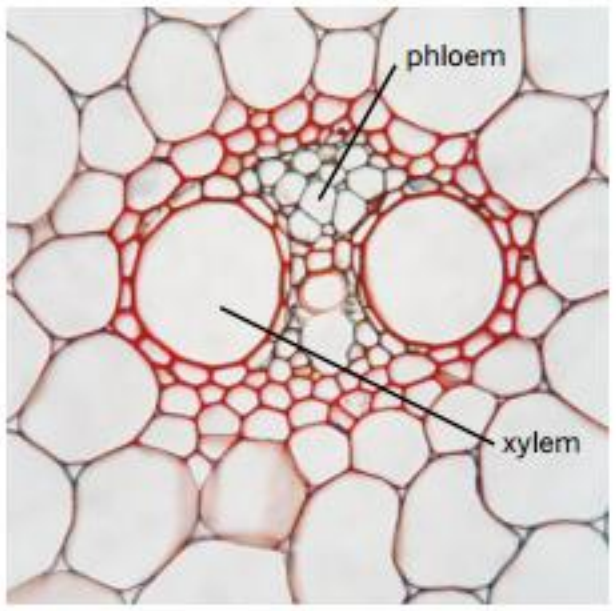


FIGURE 5.4.9 A cross-section through a stem, showing the vascular bundle containing xylem and phloem.

Tracheids

Tracheids are single, large, tapering water-filled cells that form part of the xylem tissue in all vascular plants (Figure 5.4.10). When mature, tracheids lose their nucleus and cytoplasm. This leads to cell death but creates an open structure for water to flow through. Mature tracheids have:

- cylindrical skeletons of dead cells joined to form continuous tubes, like xylem vessels
- pits and perforations in their lignified cell walls
- no nucleus or cytoplasm.

Unlike xylem vessels, tracheids are not connected end to end. Instead, their ends overlap, and water is transferred horizontally through the adjoining pits.

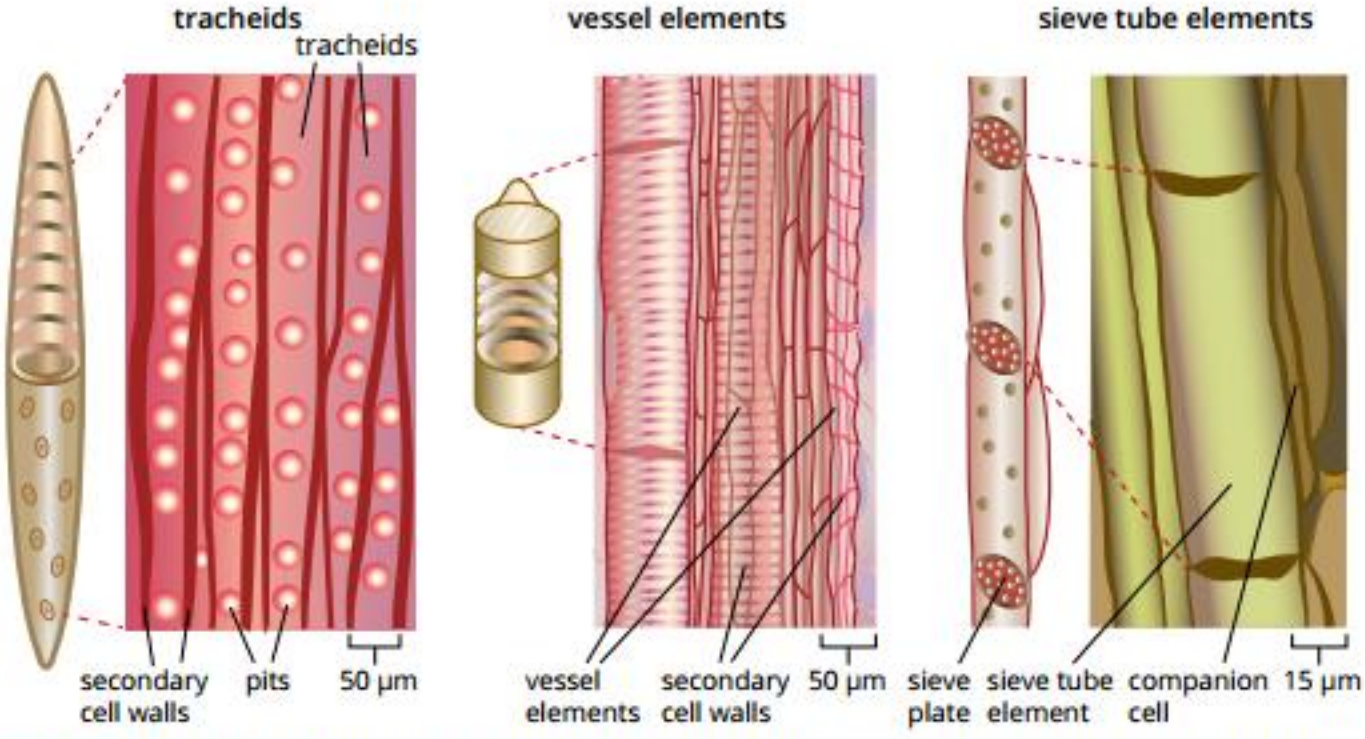


FIGURE 5.4.10 Water and mineral ions travel through tracheids and vessels in the xylem tissue. This figure shows the different forms of individual xylem elements. Tracheids exist singly and are connected through pits along their cell walls, while xylem vessels are joined end to end to form a long tube. Sieve tubes form part of the phloem.

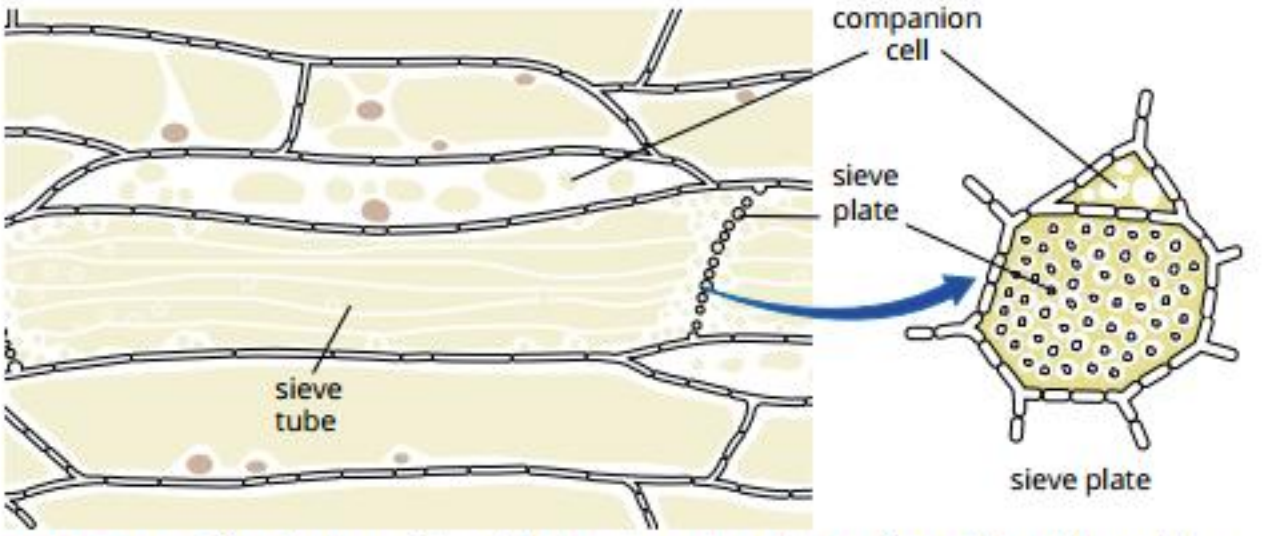


FIGURE 5.4.11 The cytoplasm of sieve tubes in phloem is continuous from cell to cell through the sieve plates. Adjacent companion cells probably enable sieve tube cells to continue functioning, since sieve tube cells do not have a nucleus.

Phloem

Phloem transports organic solutes, especially sugars such as sucrose, from the site of synthesis (leaves) to the site of use or storage (stems and roots). It is composed of sieve tubes, companion cells, parenchyma cells and sclerenchyma cells (Figure 5.4.11).

Unlike xylem vessels, mature phloem sieve tubes are living cells with no nucleus and no lignin in the cell walls. Sieve tubes form linear rows of elongated cells. Their cell walls are perforated at each end by a number of holes or pores, forming sieve plates. Strands of cytoplasm (**plasmodesmata**) pass through these perforations, connecting one cell with the next.

Sieve tube cells are usually closely associated with one or more companion cells, connected by plasmodesmata. Both sieve tube cells and companion cells have thin cell walls. But unlike sieve tube cells the companion cells retain their nucleus, which, it is thought, enables the sieve tube cells to continue functioning.

Leaves

In vascular plants a leaf is an organ composed of three distinct layers of specialised cells, or tissues (Figure 5.4.12):

- upper epidermis
- mesophyll
- lower epidermis.

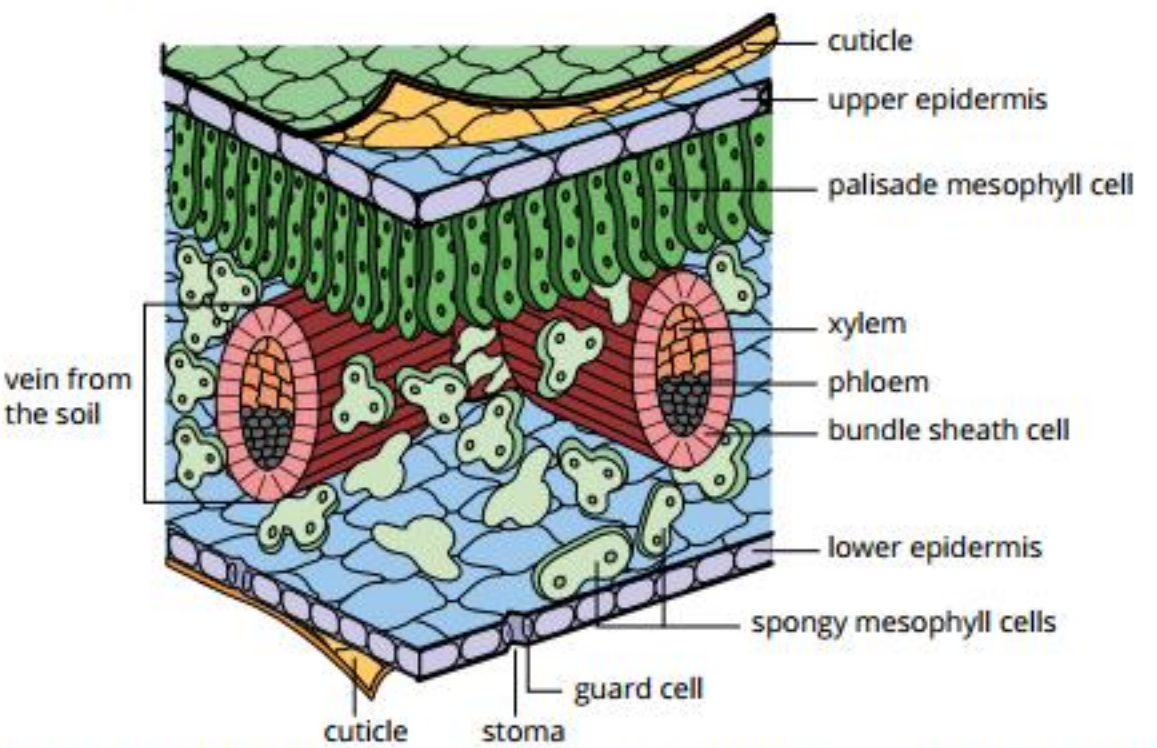


FIGURE 5.4.12 The three distinct layers of cells in leaves are the upper epidermis, the mesophyll and the lower epidermis.

The epidermis is a layer of cells covering the entire leaf. It secretes a waterproof waxy layer called the cuticle. Together the epidermis and cuticle provide a barrier that protects the cells and tissues inside the leaf and prevents excessive water loss. The epidermal cells lack chloroplasts but are transparent, allowing sunlight to reach the photosynthetic cells below.

Within the lower epidermis are stomata (pores). The stoma sits between two highly specialised epidermal cells called guard cells. When the guard cells surrounding the stomal pore fill with water, they create an opening through the epidermis and cuticle. Guard cells regulate gas exchange and water loss by changing shape, which causes the pore to open or close.

Between the epidermal layers are the mesophyll cells where photosynthesis takes place. The cells closest to the upper epidermis are the palisade mesophyll cells. These cells contain many chloroplasts and are tightly packed together. The spongy mesophyll cells below the palisade mesophyll cells are loosely packed together, with air spaces between them to allow gas exchange. These cells contain fewer chloroplasts.

The vascular tissue (xylem and phloem) is also located between the two layers of epidermal cells.

Table 5.4.1 Summarises the structure and function of the parts of leaves.

TABLE 5.4.1 The structure and function of the specialised cells and tissues of leaves

Leaf tissue	Structure	Function
cuticle	thin, waxy waterproof layer	protects the inner cells, prevents water loss and allows sunlight to penetrate
epidermis (upper and lower)	transparent and usually thin	protects the inner cells, prevents water loss and allows sunlight to penetrate for photosynthesis
epidermis and cuticle	contains guard cells surrounding stomata	regulate gas exchange and water loss by opening and closing stomata
mesophyll	palisade mesophyll; tightly packed column-shaped cells with many chloroplasts, close to upper epidermis	photosynthesis
	spongy mesophyll; loosely packed, with air spaces around the cells	allows gas exchange, including the diffusion of carbon dioxide throughout the leaf
xylem and phloem	tubular vessels	transport fluids

MOVEMENT OF WATER AND SOLUTES

Root absorption

Roots have a branched structure that increases the surface area of the roots and their capacity to absorb water and mineral ions (Figure 5.4.13).

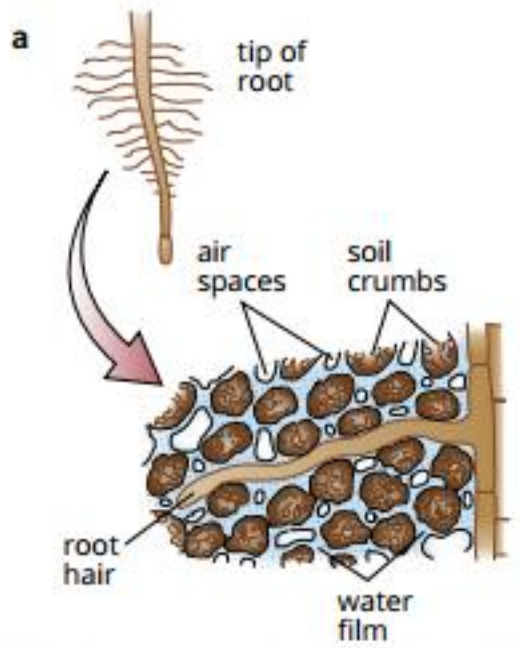


FIGURE 5.4.13 (a) Water and inorganic nutrients are absorbed by roots from soil water through many fine root hairs. (b) Root hairs on a radish seedling. The branched structure of the fine root hairs provides a greater surface area for the radish seedling to absorb water.

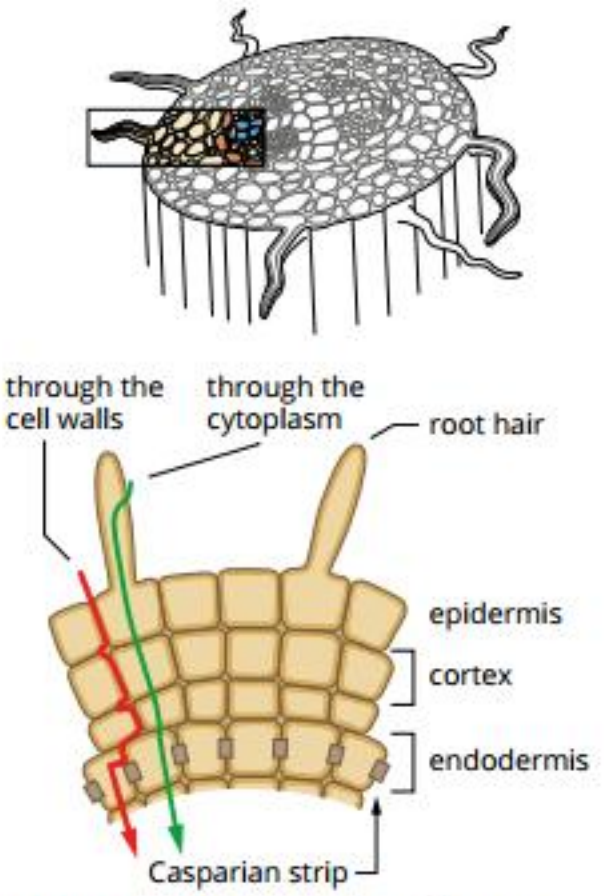


FIGURE 5.4.14 Water and mineral ions move through the roots via the extracellular pathway (red arrow) and the cytoplasmic pathway (green arrow). From the Casparian strip, water can no longer travel along the extracellular pathway and is forced into the cytoplasm before moving into the xylem.

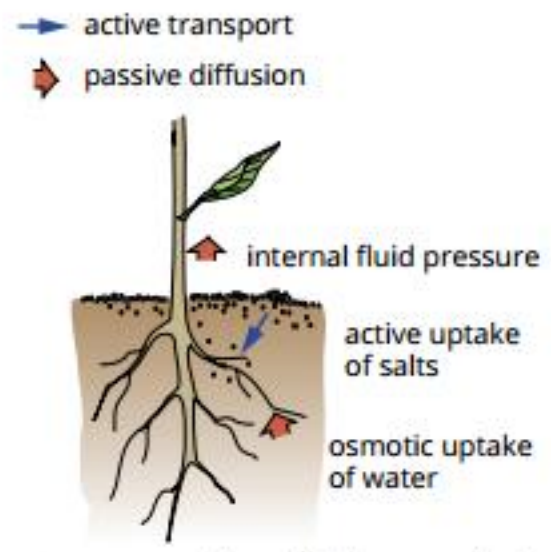


FIGURE 5.4.15 Internal fluid pressure (root pressure) in the roots of some plants causes fluid to rise through the xylem vessels.

Water pathways

There are two possible pathways for movement of water and mineral ions absorbed from the soil through the roots. These are the extracellular pathway and the cytoplasmic pathway.

In the extracellular pathway, most water and some mineral ions pass in or between cell walls (Figure 5.4.14).

In the cytoplasmic pathway, most mineral ions and some water pass through the cytoplasm of living root cells (Figure 5.4.14).

The cytoplasmic pathway involves substances entering a root hair cell by crossing the cell's plasma membrane, and then passing from cell to cell through plasmodesmata. The three types of transport that move substances across plasma membranes along the cytoplasmic pathway are as follows:

- Active transport—most dissolved mineral ions are selectively taken into roots by active transport. Proteins in the plasma membrane of root cells, specific for each ion, are used for this purpose. As a result, the concentration of ions in the vascular tissue of roots can be more than 100 times their concentration in the water of the surrounding soil.
- Osmosis—the high concentration of ions in the vascular tissues of terrestrial plants creates a very large osmotic concentration gradient. Large amounts of water move into root cells along this concentration gradient.
- Diffusion—some mineral ions such as potassium and phosphate enter the roots by diffusion. The uptake of these nutrients therefore depends on the rate of water uptake.

Entering the xylem

From either of the two pathways through the roots, water and mineral ions must then reach the xylem tissue. Between the roots and the xylem is a waterproof layer of cells that form a barrier known as the **Casparian strip**. At this barrier, water travelling through the extracellular pathway is forced into the cytoplasm. In this way the Casparian strip ensures the regulation of the substances entering the xylem.

Root pressure

In some plants, the osmotic gradient draws in so much water from the roots that it can travel up to 10 metres up the stem. This is known as **root pressure** (Figure 5.4.15). Root pressure causes the rising of sap (water and mineral ions) in spring in deciduous plants such as birch trees but it does not occur in all plants.

Transpiration

Transpiration is the passive movement of water through a plant from the roots, to its evaporation through the stomatal pores in leaves. The plant uses a small amount of water for metabolic processes, but 99% of the water absorbed by the roots is lost via transpiration.

Transpiration is a passive process: it does not require energy expenditure by the plant. It is driven by the heat energy in sunlight. Water molecules are very cohesive; that is, they have a strong tendency to stick together. When water evaporates from the cell walls of the leaf, cohesion between the water molecules remaining in the leaf draws water from nearby xylem vessels to replace the lost water (Figure 5.4.16).

In this way thousands of leaf cells, each drawing water from xylem, create a differential pressure that pulls water up xylem vessels from the roots. This continuous one-way flow of water from roots to leaves is called the **transpiration stream**. The pull of transpiration can be strong enough to draw water to the top of the tallest tree, more than 100 metres high.

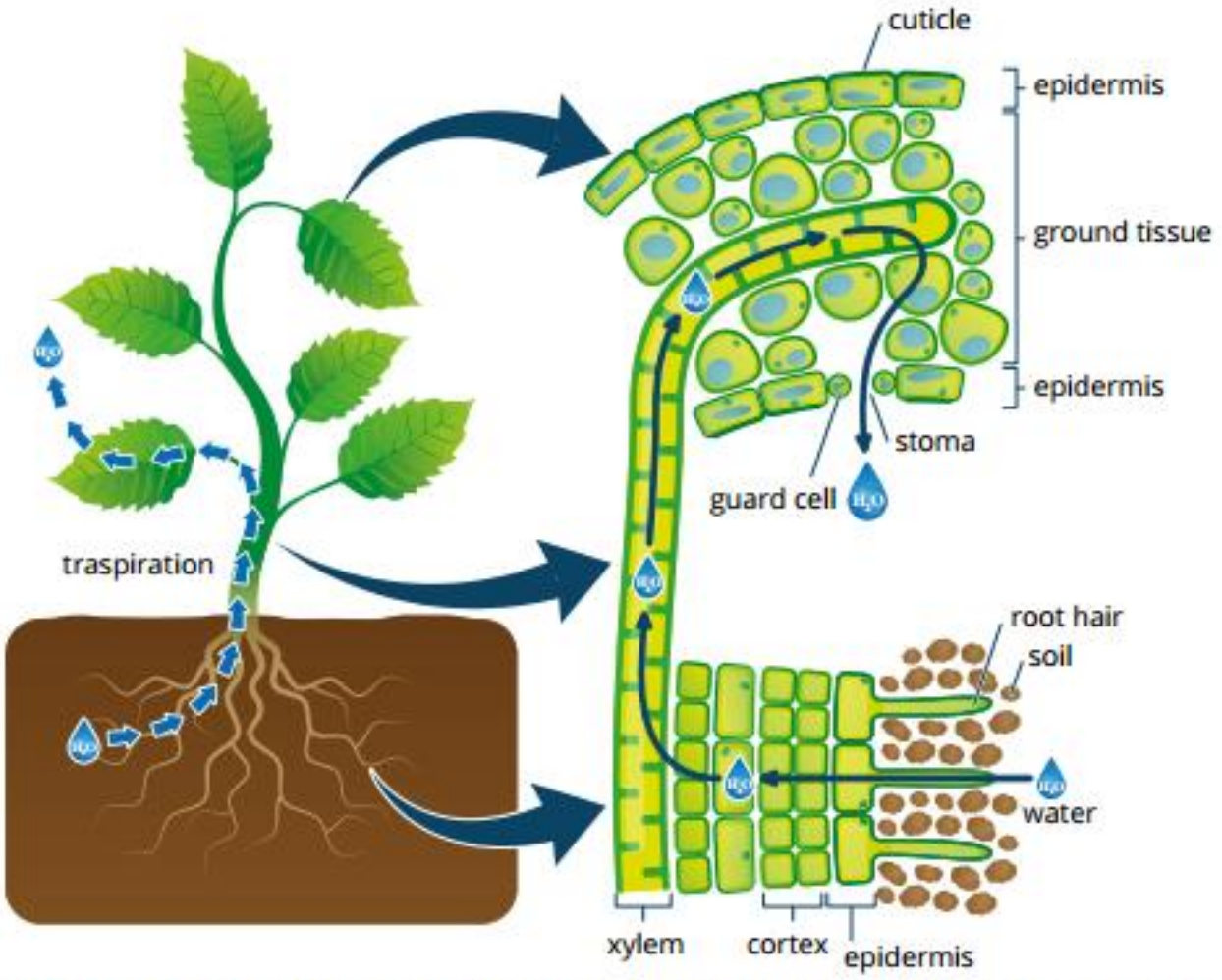


FIGURE 5.4.16 Plants draw water up from the soil through the roots and stems to the leaves. This process can occur without energy as water molecules have the property of cohesion. This means that each water molecule pulls the next up because of their attraction to each other.

> The flow of the transpiration stream does not require an intact root system. It continues when cut flowers and leafy shoots are placed in a vase of water. It also continues in the roots after the stem dies.

Although transpiration is the cause of 99% of a plant's water loss, it is vital because it enables plants to:

- absorb the water necessary for the process of photosynthesis
- transport mineral salts to leaf cells and fruits
- cool down and not become overheated.

Factors that affect transpiration rates

Water vapour is lost from leaves mainly by transpiration through open stomata. The total surface area across which transpiration takes place is related to the degree of opening of all stomata. This is by far the most important factor affecting the rate of transpiration. The greater the number of stomata and more open they are, the more surface area there is from which water can be lost (Figure 5.4.17).

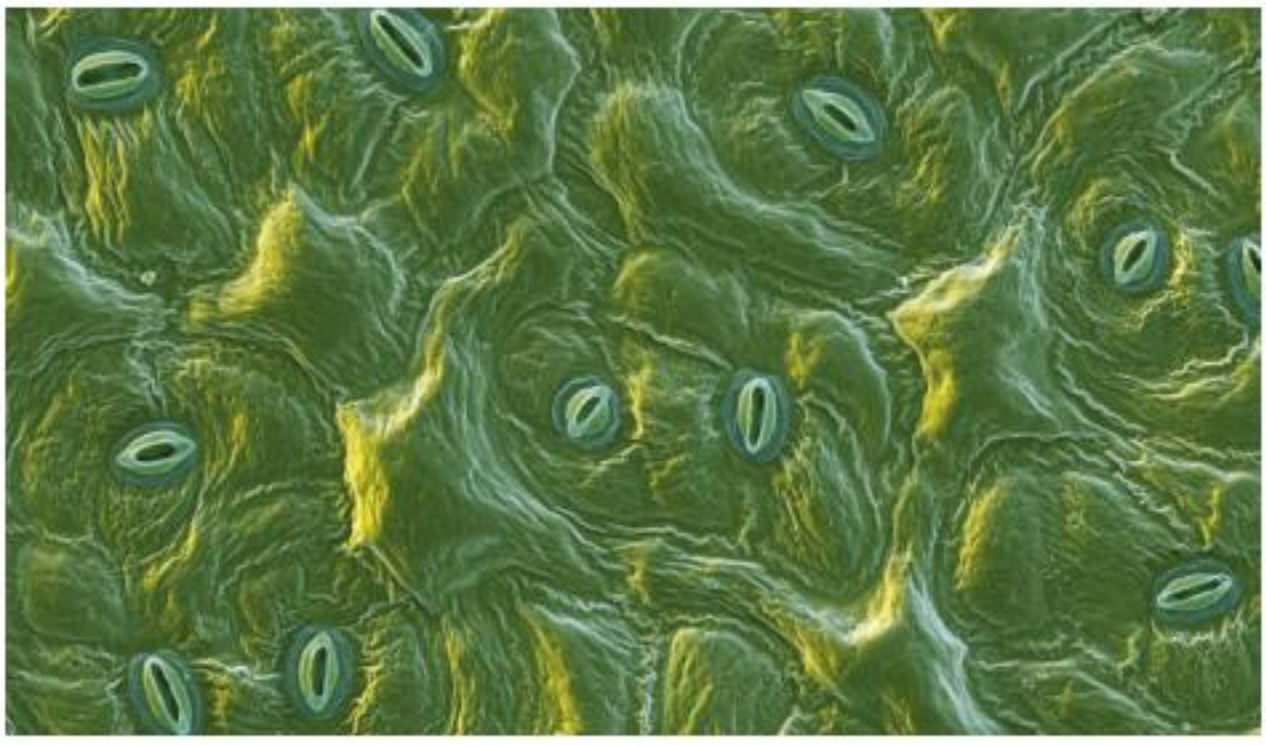


FIGURE 5.4.17 Stomata on the surface of a cabbage leaf, viewed at high magnification. Stomata regulate the exchange of gas and water between the plant and the atmosphere. The opening and closing of the stomata are controlled by two guard cells on either side of the pore. Stomata open during the day to exchange gases during photosynthesis and close at night to minimise water loss.

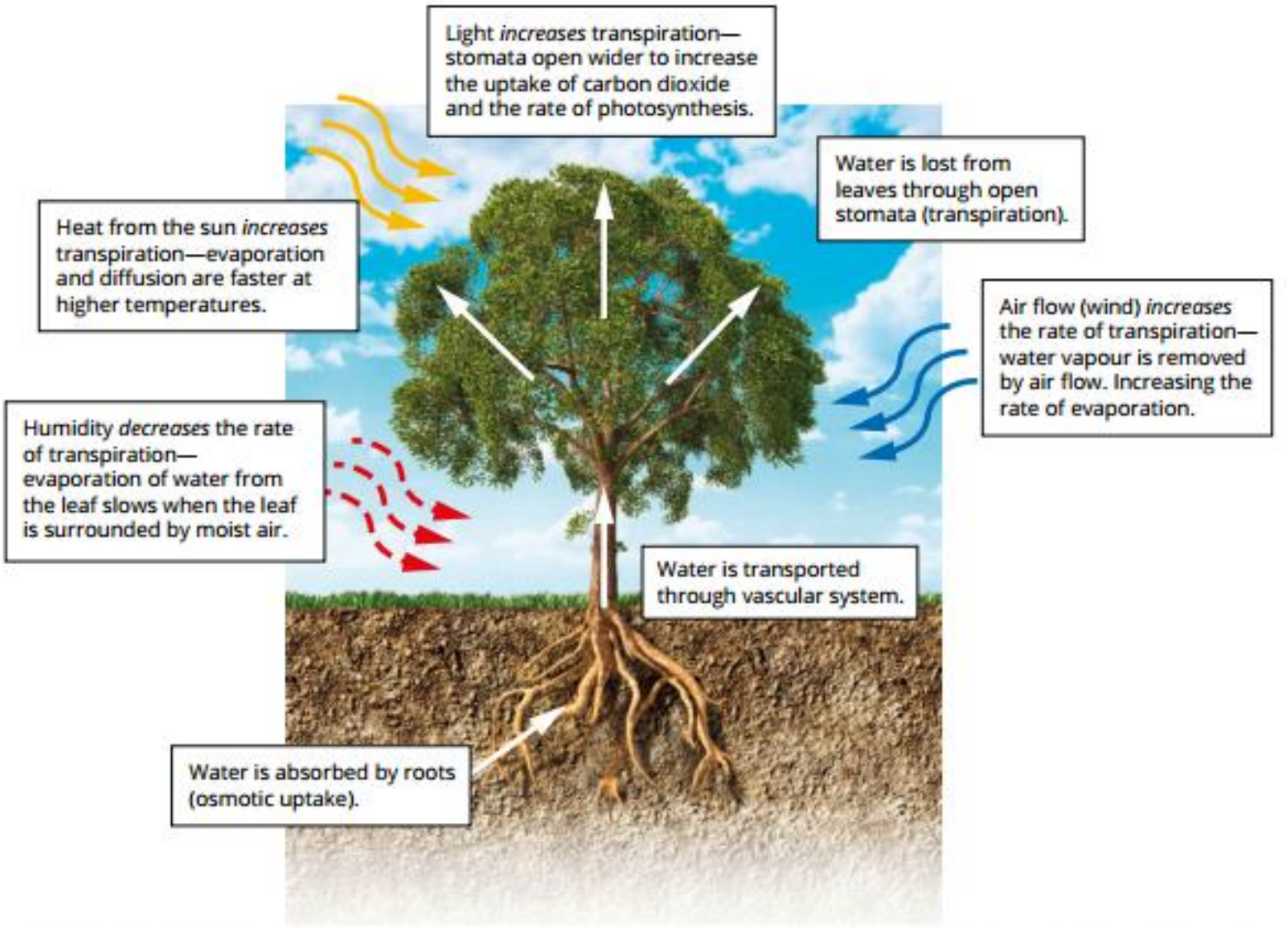

Light *increases* transpiration—stomata open wider to increase the uptake of carbon dioxide and the rate of photosynthesis.
Water is lost from leaves through open stomata (transpiration).
Heat from the sun *increases* transpiration—evaporation and diffusion are faster at higher temperatures.
Air flow (wind) *increases* the rate of transpiration—water vapour is removed by air flow. Increasing the rate of evaporation.
Humidity *decreases* the rate of transpiration—evaporation of water from the leaf slows when the leaf is surrounded by moist air.
Water is transported through vascular system.
Water is absorbed by roots (osmotic uptake).

FIGURE 5.4.18 The movement of water through the xylem vessels of vascular plants and into the atmosphere through leaf stomata, in the form of water vapour, is the process known as transpiration.

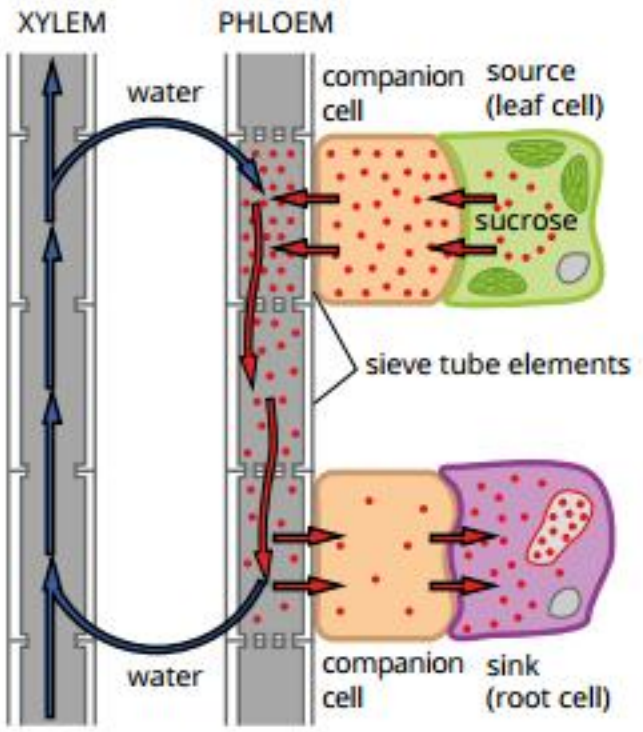


FIGURE 5.4.19 The movement of fluid through the phloem is the result of active pumping of sugars, with water flowing along an osmotic gradient. Sugars and water enter the phloem sieve tubes in leaves in this way and are translocated throughout the plant. Sugars are actively unloaded from sieve tubes where they are required.

Other factors that affect the rate of transpiration (Figure 5.4.18) include:

- humidity—transpiration rates decrease when there is a lot of water vapour in the air (i.e. a high level of humidity), because this reduces the water concentration gradient between leaf spaces and air, so fewer water molecules evaporate into the air
- temperature—transpiration rates increase as temperature increases because heat energy increases the rate of evaporation of water
- wind—air currents increase the rate of transpiration by moving water vapour away from the leaf and therefore increasing the rate of evaporation of water.

Environmental factors such as sunlight and humidity affect the rate of transpiration and, therefore, the rate of water uptake by the roots of the plant.

The rate of transpiration is low at night because it is cooler and more humid, and because stomata are usually closed.

The leaves of some plants that live in exposed conditions have developed structural features that reduce the rate of transpiration. For example, some plants have hairs on the leaf surface, which create a layer of relatively undisturbed, humid air.

Translocation: sources and sinks

The transport of organic solutes from the leaves to other tissues in the plant is known as **translocation**. Leaves produce carbohydrates in the form of sugars during photosynthesis. The non-photosynthetic tissues of the plant also need these carbohydrates and other organic compounds, such as amino acids, hormones and proteins, so these nutrients are transported from the **sources** (the leaves) to the **sinks** (regions where the nutrients are needed, such as roots, stems, flowers and fruits).

Translocation is an active process. It involves the flow of cytoplasm in sieve tubes cells driven by a pressure gradient and requires the expenditure of energy by the plant. The tissue through which these organic solutes move is the phloem, and the material that flows through it is known as phloem sap. This sap is composed of around 90% sucrose. Sucrose is a disaccharide that dissolves easily in water, making it a good transport material. It is produced in the chloroplasts of the chlorenchyma (parenchyma cells with chloroplasts) and pumped into the companion cells. From the companion cells the sucrose flows into the sieve tube cells (Figure 5.4.19). Transport in individual sieve tube cells is in one direction only, but bundles of sieve tube cells are able to transport sap in both directions: upwards to leaves and fruit, or downwards to the roots.

5.4 Review

SUMMARY

- Plants do not have organs specialised for gas exchange, but they have large surface areas. Their tissues exchange gases directly with their environment.
- Stomata are found in the epidermis of leaves and some stems. They are the main route through which gas exchange occurs in plants.
- Conditions favouring opening of stomata are abundant water, bright light and low internal carbon dioxide concentrations.
- Roots exchange gases with air in soil spaces. The crumb structure of the soil and the activities of earthworms are important in maintaining well-aerated soil.
- Most terrestrial plants, including ferns, conifers and flowering plants, have vascular tissues (xylem and phloem) that are specialised for transporting fluid.
- The vascular tissues are:
  - xylem, which carries water and mineral ions from roots to leaves
  - phloem, which carries sugars and other organic molecules from leaves to roots.
- Xylem vessels:
  - are the skeletons of dead elongated cells
  - have perforations at each end
  - are joined end to end to form continuous tubes and allow the flow of fluid
  - have pits (thinner areas) in the side walls that enable the movement of substances into and out of the adjacent companion cells.
- Xylem tracheids:
  - like xylem vessels are dead and have pits in their lignified cell walls and have no nucleus or cytoplasm
  - unlike xylem vessels, are not connected end to end; their ends overlap and water is transferred horizontally through the adjoining pits.
- Water and inorganic nutrients (mineral ions) are absorbed by the root hairs from the soil by one of two pathways:
  - extracellular pathway
  - cytoplasmic pathway.
- Water and mineral ions are transported through xylem vessels as sap. This transportation occurs in one direction only: from roots to leaves.
- Transpiration is the evaporation of water from stomata in leaves. It is a passive process (driven by energy from sunlight) that also draws water up from the roots, through the xylem, following what is known as the transpiration stream.
- The rate of transpiration is affected by:
  - the number of stomata and their degree of opening
  - temperature
  - humidity
  - wind.
- Translocation is the transport of organic materials from the leaves to the roots, stem, flowers and fruits of the plant, through the sieve tube cells and companion cells of the phloem tissue.
- Translocation is an active process and requires an expenditure of energy by the plant.

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5.4 Review *continued*

KEY QUESTIONS

Describe

1 State the two types of vascular tissue in plants and their functions.

2 Recall two possible pathways for the movement of water and mineral ions, absorbed from the soil, through the roots.

3 Identify one difference between how plants and how animals exchange gas with their environment.

4 Name the cells that are responsible for allowing gases to move into or out of plant leaves.

5 Name another way that plants can exchange gases with their environments.

Apply

6 Explain how it is possible for the tallest trees to transport water from their roots to their uppermost branches, sometimes more than 100 metres high.

7 Explain why most plants do not need specialised gas exchange organs.

8 Use labelled diagrams to show how stomata open and close.

9 What mechanisms might a plant adopt to reduce water loss to transpiration?

10 Determine whether each of the following environmental factors increases or decreases transpiration rates in plants.

- **a** high temperature
- **b** high humidity
- **c** darkness
- **d** strong wind

Analyse

11 Predict what will happen to gas exchange in a pot plant if you over water it.

12 Guttation is the appearance of small water droplets along the margins of leaves, caused by water being forced out of the leaf. Identify the source of the water droplets.

13 Construct a table to compare the structure and function of phloem and xylem vessels. Consider the:

- structure of cells
- properties of cells
- substances that are transported
- direction in which substances are transported
- source of energy for transport.

14 A cactus plant does not open its stomata until the evening. Identify the traditional environment of cacti and hence, infer why this might be so.

15 Year 11 Biology students were investigating the effect of air movement on the rate of transpiration of celery. They set up 3 beakers for each treatment. 100 mL water and a celery stalk were added to each beaker. After 2 hours, the volume left in the beaker was measured. The results are in Table 1.

TABLE 1 Volume of water left in beaker after 2 hours under different air movement conditions.

Treatment	Trial 1	Trial 2	Trial 3	Average
on bench	56	54	51	
under fan	22	23	19	
in glass cupboard	73	74	78	

- **a** Determine the average for each treatment.
- **b** Interpret what these results are telling the students.

Chapter review

05

KEY TERMS

aerobic
air sac
alveoli (sing. alveolus)
ammonia
amylase
antidiuretic hormone (ADH)
aorta
arteriole
artery
atrium (pl. atria)
bile
bladder
Bowman's capsule
bronchi (sing. bronchus)
caecum
capillary
carnivore
Casparian strip
cellulose
cellulase
chemical digestion
cilia (sing. cilium)
closed circulatory system
connective tissue
coronary circulation
digestion
digestive enzyme
egestion
epiglottis
epithelium
excretion
exhalation
extracellular digestion
filtration
gall bladder
glomerulus
glycogen
guard cell
haemoglobin
haemolymph
herbivore
hydrolysis
immune system
inhalation
interstitial space
intracellular digestion
lacteal
larynx
lenticel
lignin
lipase
liver
loop of Henle
lumen
lymph
metabolism
microvilli (sing. microvillus)
myoglobin
nephron
nitrogenous waste
omnivore
open circulatory system
oxygen-carrying capacity
oxyhaemoglobin
partial pressure
peristalsis
pharynx
phloem
plasmodesmata
protease
pulmonary artery
pulmonary vein
reabsorption
root hair
root pressure
rumen
secretion
sink
source
starch
stomata (sing. stoma)
tidal volume
trachea
tracheid
translocation
transpiration
transpiration stream
turgor
urea
ureter
urethra
uric acid
valve
vascular bundle
vascular plant
vascular tissue
vein
ventilation
ventricle
venule
villi (sing. villus)
vital capacity
xylem
xylem vessel

KEY QUESTIONS

Describe

1 Recall how oxygen and carbon dioxide can move freely through the respiratory surface of animals by:
A active transport **B** diffusion
C osmosis **D** facilitated diffusion

2 Identify the characteristic the mammalian respiratory surface has to make it effective.
A has a low surface area-to-volume ratio
B has the ability to ventilate itself
C has a rich blood supply
D does not need to be moist

3 Recall the correct way that blood flows around the human heart and lungs:
A right atrium – right ventricle – lungs – left atrium – left ventricle
B left atrium – left ventricle – lungs – right atrium – right ventricle
C right atrium – left atrium – lungs – left ventricle – right ventricle
D right ventricle – right atrium – lungs – left ventricle – left atrium

4 Identify the blood vessel through which the majority of gas exchange occurs:
A artery **B** vena cava
C capillary **D** aorta

Apply

5 Determine when there would be a net movement of oxygen into a plant:
A all the time
B during the day
C when the rate of gas produced in cellular respiration is greater than the rate of gas used in photosynthesis
D when the rate of gas used in cellular respiration is less than the rate of gas produced in photosynthesis

6 In autumn, the leaves of deciduous trees change colour and eventually fall. The change in colour is due to the movement of nutrients out of the leaves for storage. Recognise what is involved:
A xylem and phloem
B only the xylem
C only the phloem
D diffusion

7 Determine which of the following statements relating to fermentation in herbivores is true.

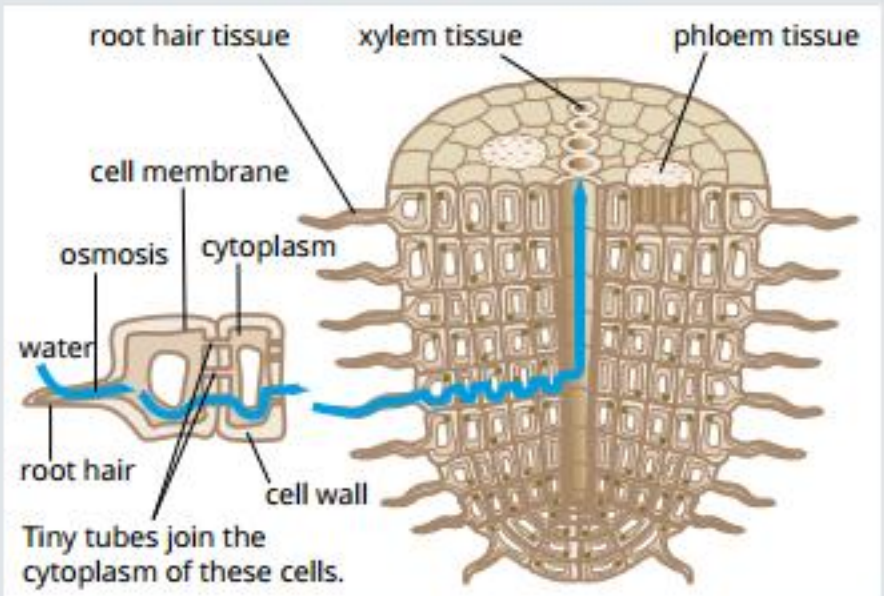
A In foregut fermenters, cellulose is digested in the caecum.

B Fermentation in the gut requires oxygen.

C The rumen is located between the oesophagus and the stomach.

D All native Australian mammals are hindgut fermenters.

8 Summarise the pathway of water absorption shown below in the diagram of plant root tissue.



9 Consider some of the features that plants living in harsh environments, such as deserts, have developed to achieve water transport.

10 **a** Identify which side of the heart has thicker muscular tissue.

b Explain why.

11 Describe the action of ADH on the collecting ducts of the nephron.

12 Recognise why vignerons (grape growers) sometimes remove the bark and stem from bunches of grapes whilst the grapes are turning colour.

Analyse

13 Coeliac disease causes the destruction of the villi cells. Assess which one of the following is most likely to happen to people with coeliac disease.

A damage in the oesophagus caused by increase in acid reflux

B incomplete digestion of proteins

C increased levels of glucose in blood

D poor absorption of calcium

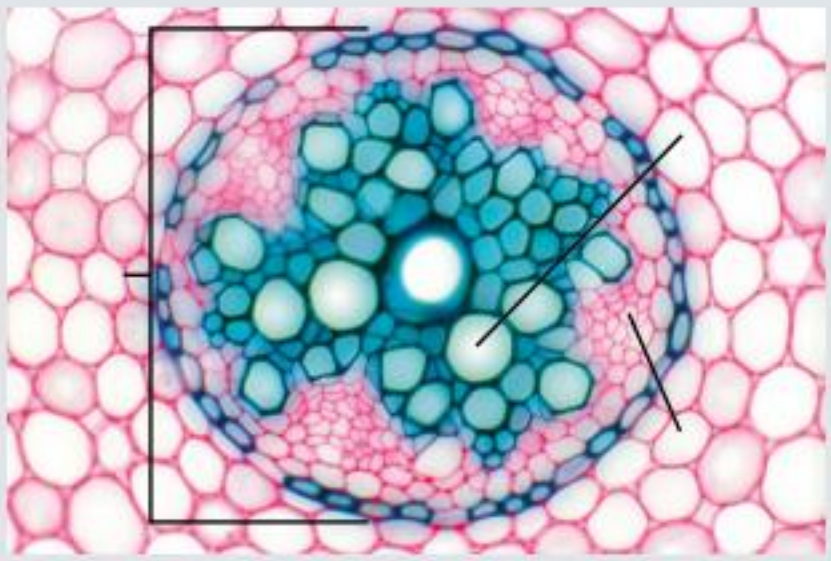
14 Infer which process and location in the kidney each of these scenarios will affect. Justify your response.

a You drink a large amount of water.

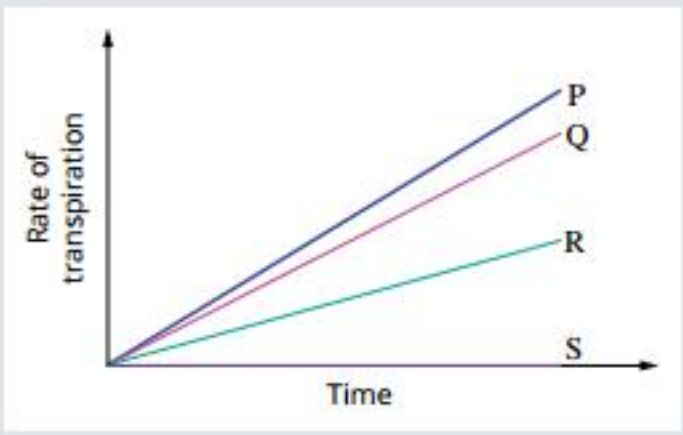
b You play vigorous sport on a hot day.

c Your blood pressure becomes very low.

15 Analyse the following cross-section of the root of a buttercup plant viewed under a light microscope. Identify the parts of the root that are shown by the two arrows and the large bracket.



16 An experiment was conducted to determine the effects of applying a sticky gel onto a leaf on the rate of transpiration. The graph below shows the results of the experiment.



Differentiate each line labelled on the graph by matching to the correct experimental condition from the list below.

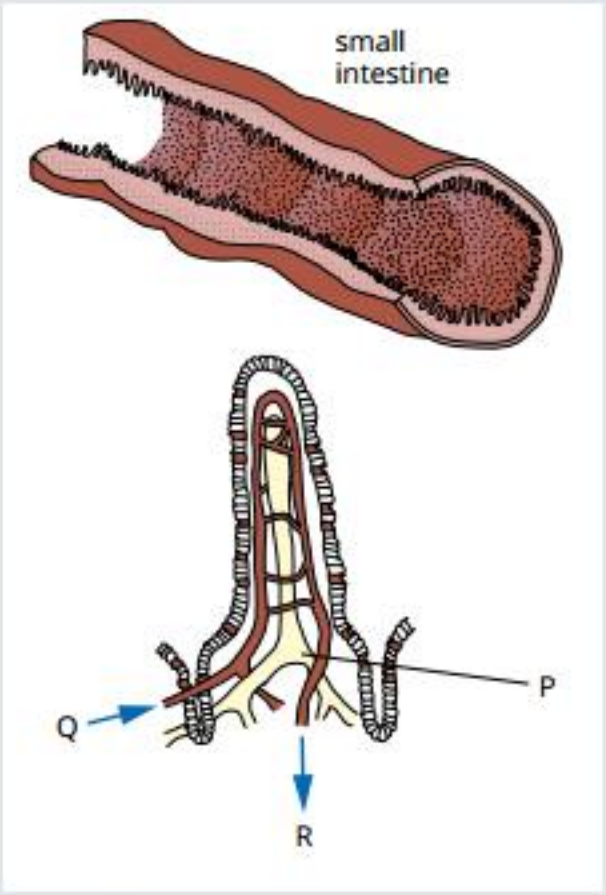
a no gel applied

b gel applied to the lower side of the leaves

c gel applied to the upper side of the leaves

d gel applied to the lower side and the upper side of the leaves

17 The following diagrams show a cross-section through the small intestine and a longitudinal section through a villus.

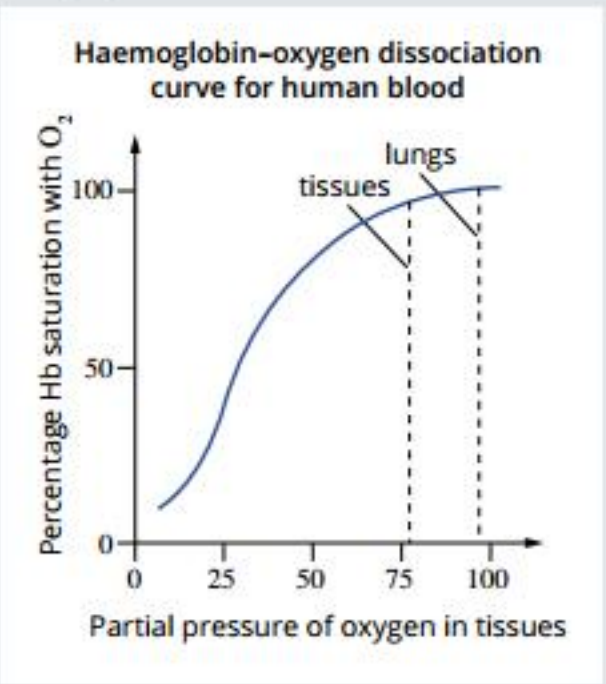


a Determine three ways in which the structure of the small intestine is related to its function of absorbing products of digeston.

b Classify structure P and state its function.

c The arrows in the second diagram indicate the direction of blood flow. Predict how the composition of blood entering from Q would be different from blood leaving R.

18 Analyse the information in the following graph. Explore the relationship between oxygen, haemoglobin and the circulatory system.



19 The table shows the partial pressures of gases at a site of gas exchange in the human body.

Location A	P_{O_2} = 102 mmHg P_{CO_2} = 40 mmHg
Location B	P_{O_2} = 40 mmHg P_{CO_2} = 45 mmHg

Derive in which direction the gases move between locations A and B.

20 A gas mixture contains 15% oxygen, 40% nitrogen and 45% helium. If the total pressure of the gas mixture is 800 mmHg, determine the partial pressures of the three gas components.

21 The teeth of a herbivore and the teeth of a carnivore are different.

a Describe what teeth you are likely to see in the mouth of each animal.

b Explain why these teeth would be present.

22 Consider the life of an Australian fur seal, (*Arctocephalus pusillus doriferus*). It spends some time on land but generally most of its time in the water. It will suckle its young on land. Whilst it is not very agile on land, it is very graceful in the water.

a Classify this fur seal as an omnivore, carnivore or herbivore.

b Predict the type of food that this animal might eat based on your classification.

c Infer the type of breathing mechanism that this animal has.

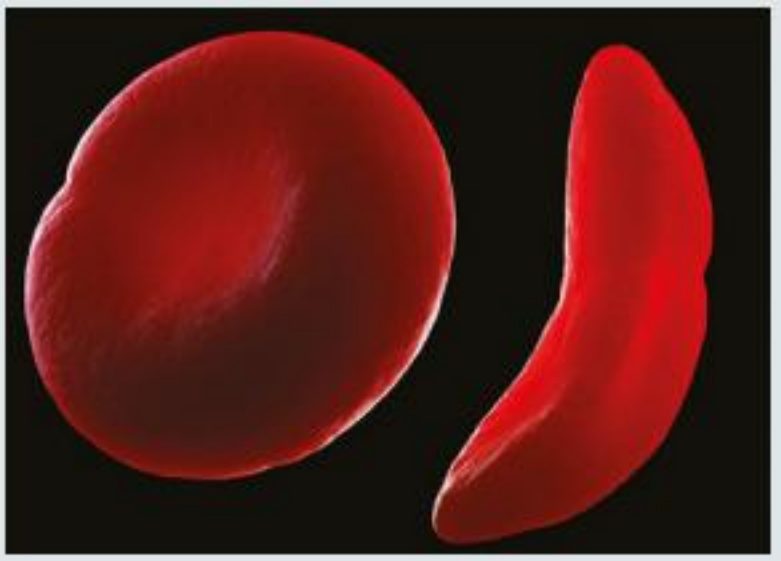
d Assess the lifestyle of the Australian fur seal and suggest whether this breathing mechanism is actually the most efficient for this animal.

23 Compare and contrast the lymphatic system and the circulatory system.

Interpret

24 Sickle-cell anaemia is a red blood cell disorder. People with the disease have abnormal haemoglobin in their red blood cells. As a consequence, the red blood cells become sickle shaped and inflexible.

Based on your understanding of the functions of red blood cells, discuss the effects of having sickle-shaped red blood cells on the functioning of the circulatory system.



25 Research three specialised cells in the respiratory system and state their functions in relation to gas exchange.

26 Devise an experiment you could carry out to determine if caffeine is a diuretic.

27 Year 11 Biology students were investigating the transpiration stream in celery. Their research question was 'Did the type of fluid that the celery sits in, have an impact on the transpiration stream of the celery?'

- **a** Determine the independent variable for the investigation.
- **b** Suggest a control that could be used in this experiment.
- **c** Propose a hypothesis for this investigation.
- **d** Design a method for this investigation.
- **e** If the students conducted the investigation, determine the results that would support their hypothesis.
- **f** The students put their celery in different areas around the room. What effect would this have on the results and the investigation?

28 The following table shows the relative concentrations of urea, glucose, amino acids, salts and proteins in the primary filtrate and urine of a young football player as a percentage of the concentration in blood plasma. The person's doctor asked for this urine sample, so that a baseline could be constructed. This was to ensure that during the season, any variations could be monitored and dealt with. The player is to give one sample every month throughout the season.

TABLE 1: First sample—before the season begins

Substance	Primary filtrate (%)	Urine (%)
urea	100	700
glucose	100	0
amino acids	100	0
salts	100	200
proteins	0	0

- **a** Discuss why there are no proteins within the sample.
- **b** Propose why there were no amino acids or glucose molecules in the urine sample.
- **c** Comment on the amount of urea (%) in the urine.
- **d** During the season, the player receives a heavy knock to their kidneys. Discuss how this would affect the urine sample and propose why these changes would occur.

Data analysis

DATA SET 1

The information below applies to Questions 1–7.

An experiment was conducted by a group of Year 11 Biology students to investigate how the body responds to increased energy requirements. The students investigated the research question 'Does increasing the effort of exercise increase the heart rate of a person?'

The raw data for this experiment is presented in Table 1.

TABLE 1 Heart rate (beats per 15 seconds) for each activity

Person	Resting	Gentle walking	Brisk walking	Running
A	20	27	33	42
B	26	31	36	45
C	23	29	32	38
D	20	26	35	41
E	24	34	41	45
F	19	24	29	34
G	23	27	32	36
H	26	32	38	43
I	22	29	34	39
J	22	31	35	39
K	19	21	24	26
L	22	28	33	41

Independent variable: effort of exercise

Dependent variable: heart rate (beats per minute)

Aim: To investigate how the heart rate changes with increased effort of exercise.

Hypothesis: If the effort of exercise increases, then the heart rate will increase.

Question 1 (1 mark)

Comment on the data set.

Question 2 (1 mark)

What mathematical technique would the group use to ensure that any unusual or anomalous result has minimal impact on the overall results?

Question 3 (2 marks)

a Calculate the average heart beats per minute for each activity.

b Construct a graph to show these results.

Question 4 (1 mark)

What is the relationship between exercise effort and heart rate?

Question 5 (1 mark)

Deduce which activity type had the greatest heart rate.

Question 6 (2 marks)

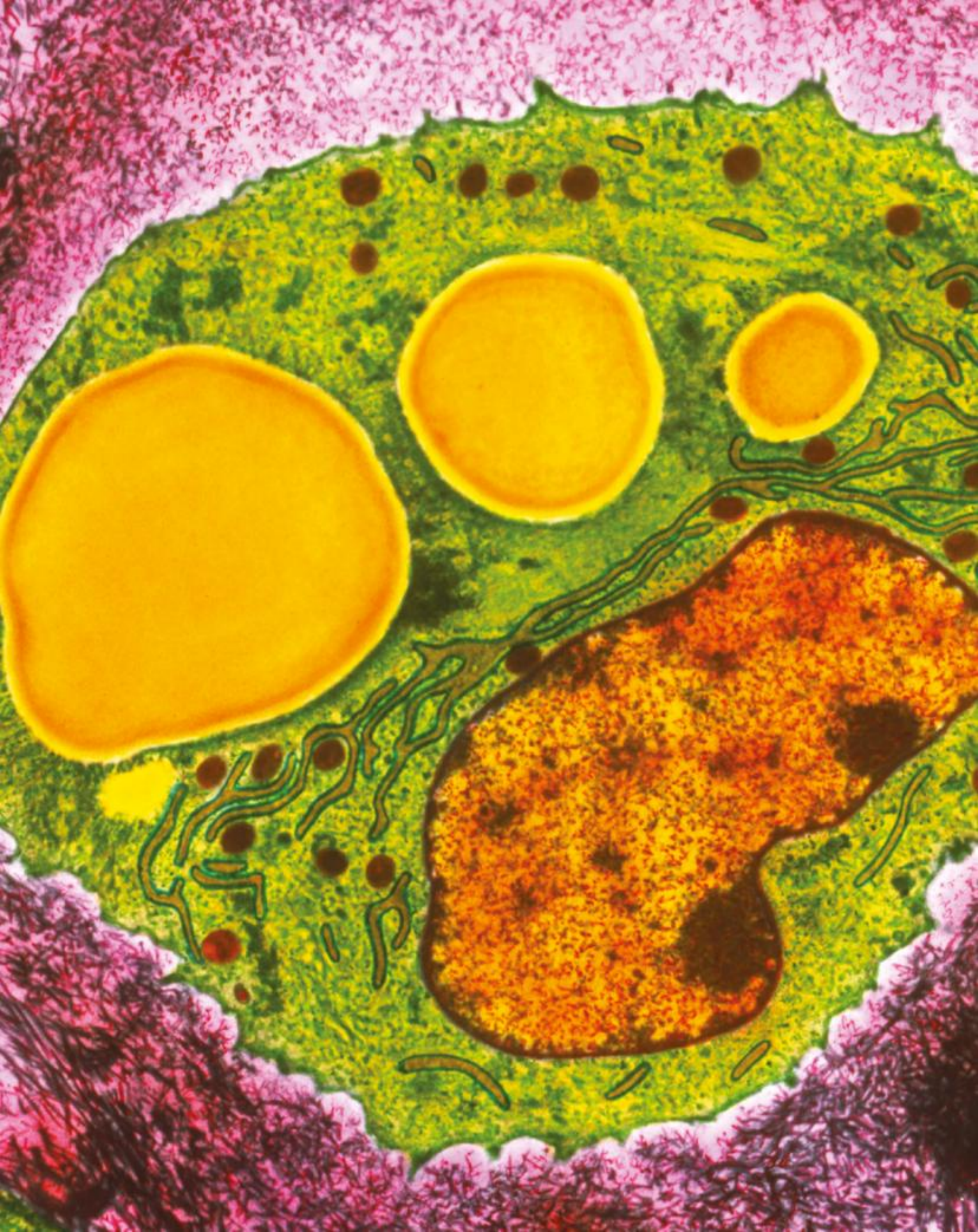
Infer from your results, why greater exercise effort resulted in a higher heart rate.

Question 7 (2 marks)

Draw conclusions from the data, about the heart rates of people in the following situations and explain why you came to those conclusions:

a person lying on the sand at the beach

b swimmer in the 400 m freestyle final at their school swimming carnival



REVIEW QUESTIONS

Topic 1: Cells as the basis of life

Multiple-choice questions

1 Select which of the following would not be visible under a light microscope.

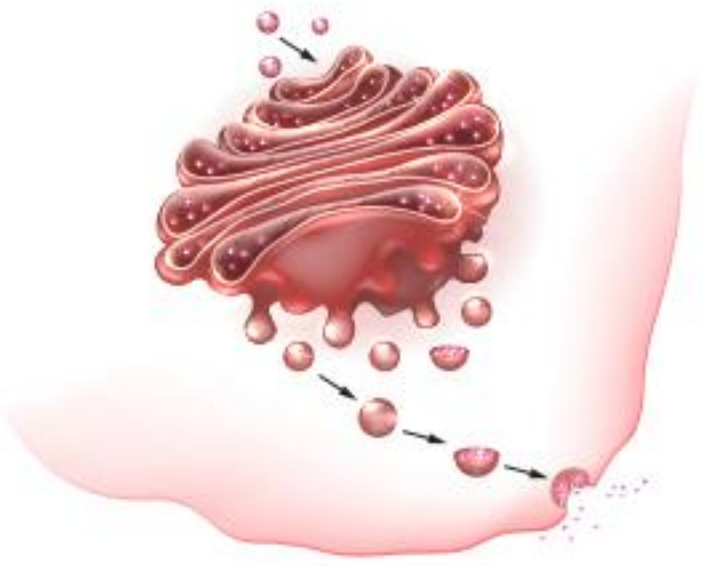
A nucleus
B chloroplast
C vacuole
D ribosome

2 Identify which of the following features is/are present in mitochondria but not in chloroplasts.

I circular DNA
II ribosomes
III outer and inner membranes
IV cristae

A I, II and III only
B II, III and IV only
C I only
D IV only

3 In the following diagram, macromolecules are being transported to the exterior of a cell.



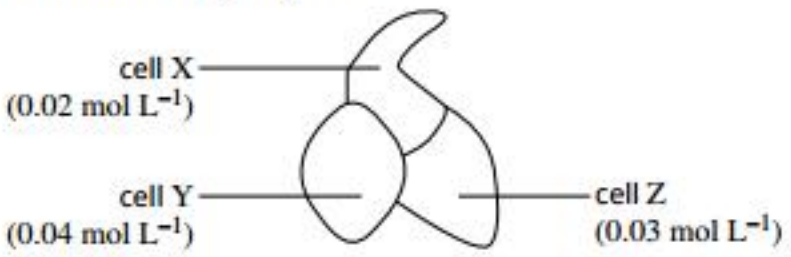
Select the name of this process.

A exocytosis
B pinocytosis
C endocytosis
D phagocytosis

4 Identify the major factor that determines whether entry to the cell by a substance will be by endocytosis.

A size of the molecules being moved across the membrane
B polarity of the substance
C water solubility of the substance
D concentration gradient of the substance between the inside and outside of the cell

5 Three cells X, Y and Z containing different solute concentrations were placed next to each other, as shown in the following diagram.



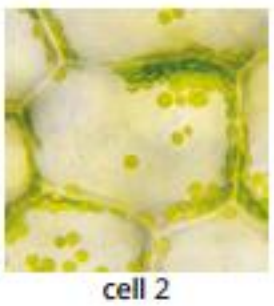
Predict the direction in which osmosis will occur.

A from X to Y only
B from X to Y, X to Z and Z to Y
C from Y to Z only
D from Y to Z and Z to X

6 Classify each cell below as eukaryotic or prokaryotic and then select the option that only includes eukaryotes.

A cell 1, cell 2, cell 3 and cell 4
B cell 1, cell 2, cell 3 and cell 5
C cell 2, cell 3, cell 4 and cell 5
D cell 1, cell 2, cell 4 and cell 5

cell 1


cell 2

cell 3

cell 4

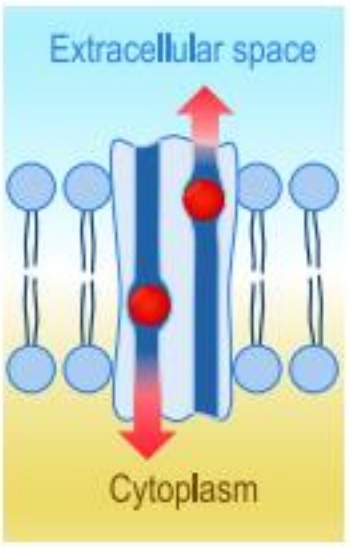
cell 5

7 Recall which of the following increases the surface area of membranes of cells without changing cell volume.

I increasing the cell size
II cell compartmentalisation
III a flattened shape
IV plasma membrane extension

A I and II only
B II and III only
C I, II and III only
D II, III and IV only

8 Identify which type of protein channel is shown in the illustration below.



A symporter
B antiporter
C uniporter
D aquaporin

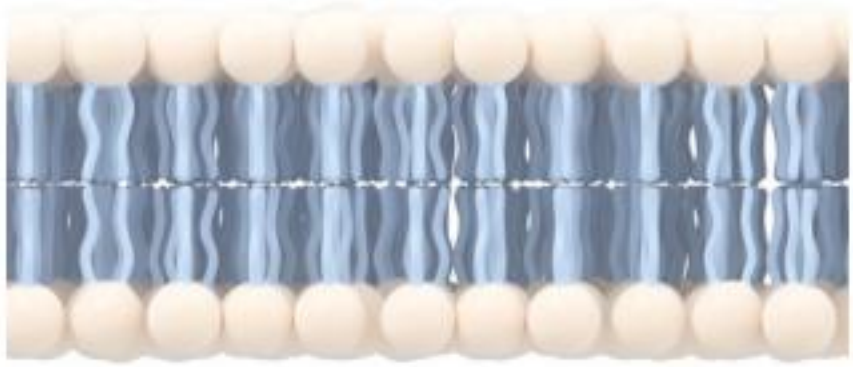
9 *POU5F1* is a gene that codes for a protein called OCT4. The concentration of OCT4 has been shown to be reduced in cells that have undergone differentiation.

Consider the diagram below of the development of a human embryo. Early developmental stages are shown in light grey, and development and differentiation progress until complete at the end of the period, shown in dark grey.

Analyse the diagram and determine when the highest levels of OCT4 would be expected.

A upper limbs at six weeks
B heart at seven weeks
C ears at eight weeks
D teeth at nine weeks

10 The diagram below shows a section of cell membrane and the spaces between the molecules. These spaces are formed as a result of:



A protein channels
B cholesterol
C saturated fatty acids
D unsaturated fatty acids

11 Most amino acids are small and polar. How do they enter a cell?

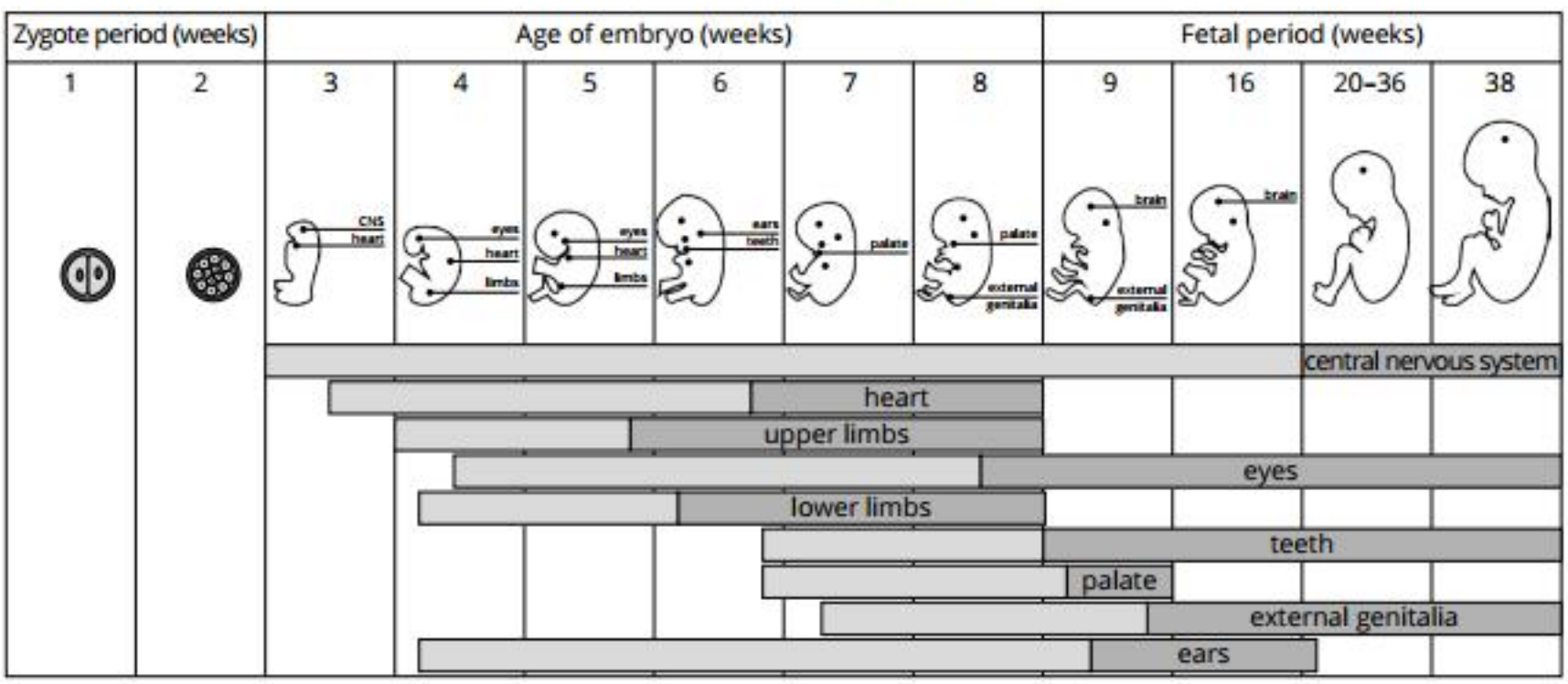
A through the phospholipid bilayer
B through a protein channel or carrier
C through phagocytosis
D through pinocytosis

12 The building blocks of proteins are:

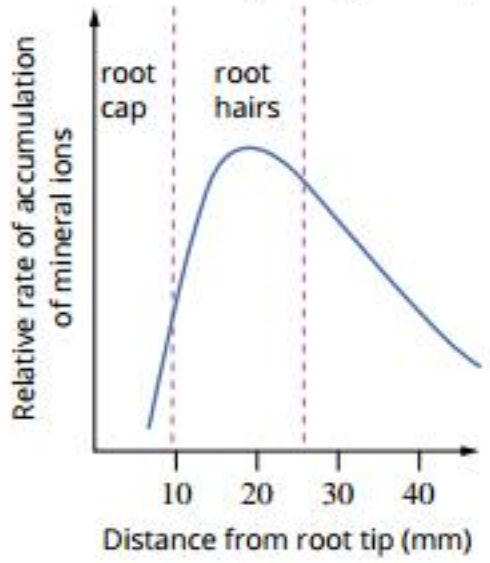
A monosaccharides
B fatty acids
C amino acids
D nucleic acids

13 Cells with the greatest potential to become the greatest variety of cells are those that are:

A unipotent
B multipotent
C totipotent
D pluripotent



14 The following graph shows the relative rate of accumulation of minerals in the root of a plant at different distances from the growing root tip.



Identify the conclusion that can be drawn from the data.

A Active transport causes ions to accumulate in the root.

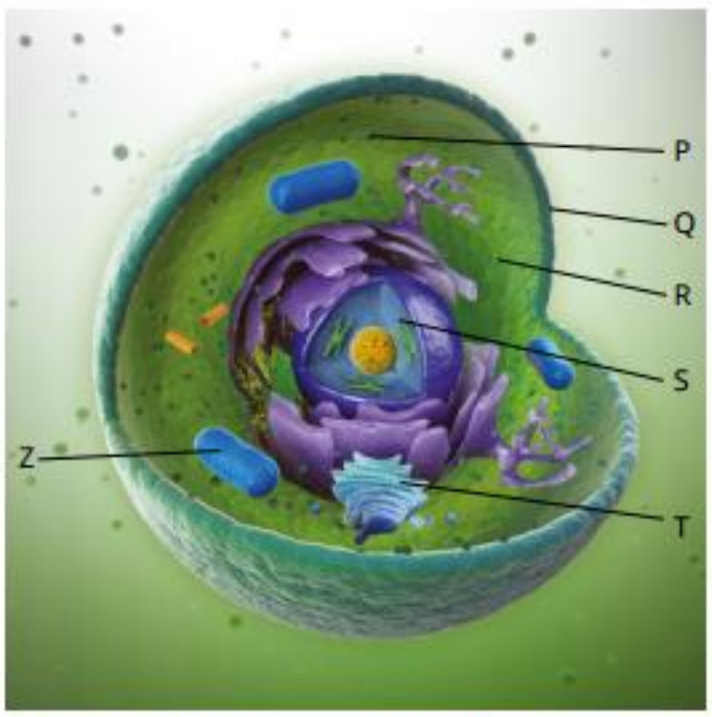
B At larger distances from the tip all the minerals are lost from the root.

C A large surface area is necessary for mineral accumulation.

D Mineral accumulation is greatest in the part of the root with root hairs.

Short-answer questions

15 The following is a three-dimensional image of an animal cell.



a Identify structures P, Q, R, S and T.

b Describe a key feature of structure Q. Explain how this feature relates to its function.

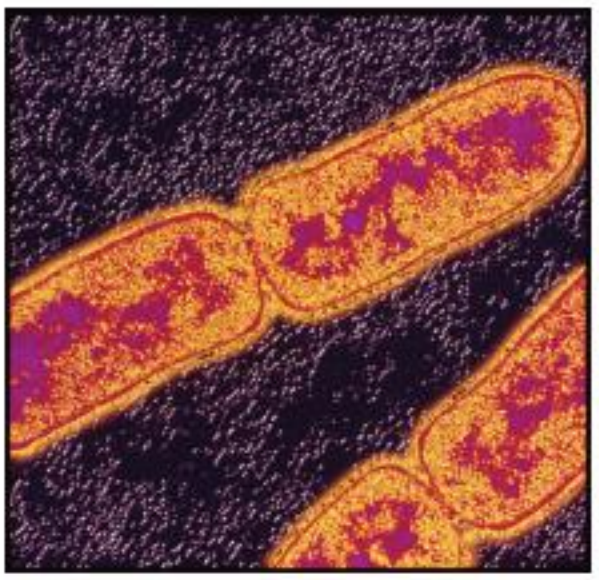
A transmission electron micrograph of structure Z is shown in the following figure.

c Identify structure Z.

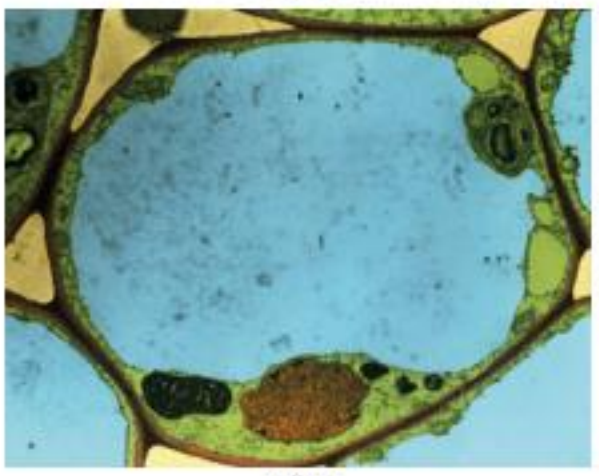
d Explain how the structure of Z assists the cell to perform its function.

e Explain how oxygen from the blood eventually enters structure Z.

16 The following images show the transmission electron micrographs of two cells, P and Q.



cell P



cell Q

a Identify the image that shows a prokaryote. Justify your answer.

b Draw an arrow and label the structures where DNA can be found in each picture.

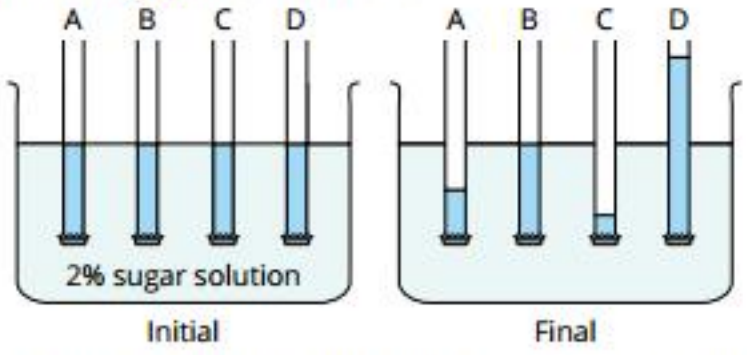
c Name two features in cell Q that are not visible in cell P.

17 **a** What is meant by the term 'compartmentalisation' in reference to eukaryotic cells?

b How does compartmentalisation benefit eukaryotic cells?

18 Four tubes contain solutions with different sugar concentrations. One end of each tube is covered by a semipermeable membrane. The tubes are placed in a tank containing a 2% sugar solution and are left until the fluid levels are stable, as shown in the diagram.

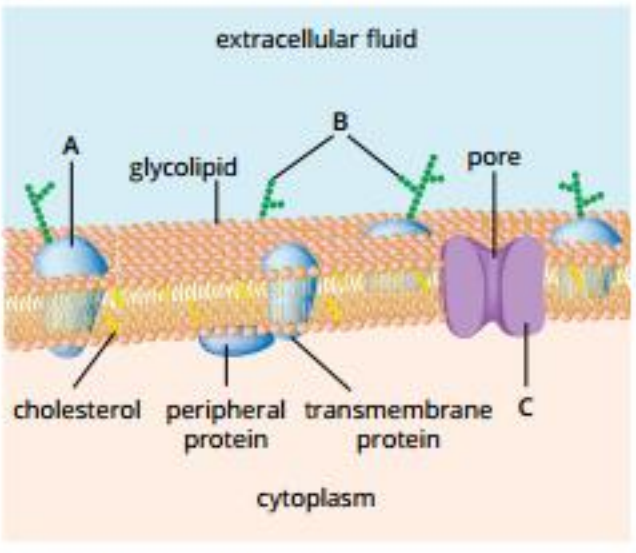
Conclude the original concentrations of solutions A, B, C and D. Justify your conclusions.



19 **a** Describe a similarity and a difference between simple diffusion and facilitated diffusion.

b A student describing to their parent what they had been studying in class stated that they had been learning about transport across cell membranes. However, they were quite confused and explained that the process of endocytosis was a kind of facilitated diffusion. Compare endocytosis and facilitated diffusion to explain why the student was incorrect.

20 The following diagram illustrates the structure of a cell membrane.



a Identify molecules A and B.

b **i** Name structure C.

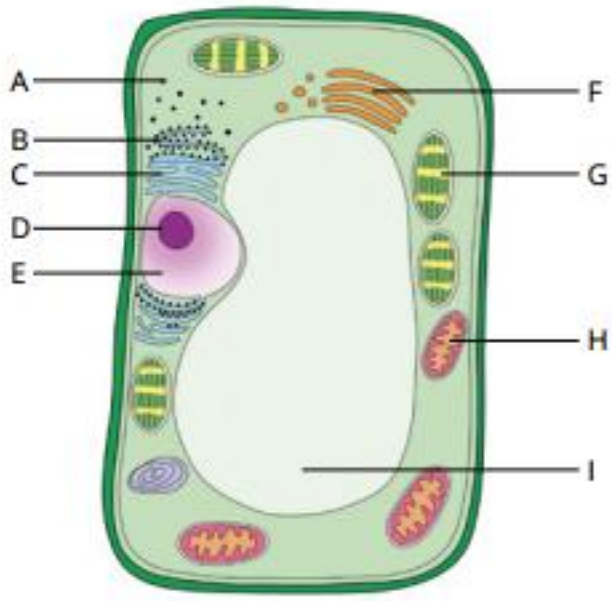
ii Describe the function of structure C.

c Explain the function of cholesterol in the cell membrane.

21 Many of the ribosomes found in a cell are attached to the endoplasmic reticulum, forming rough endoplasmic reticulum.

Discuss how this arrangement benefits the functioning of a cell.

22 **a** The following diagram shows a eukaryotic cell.



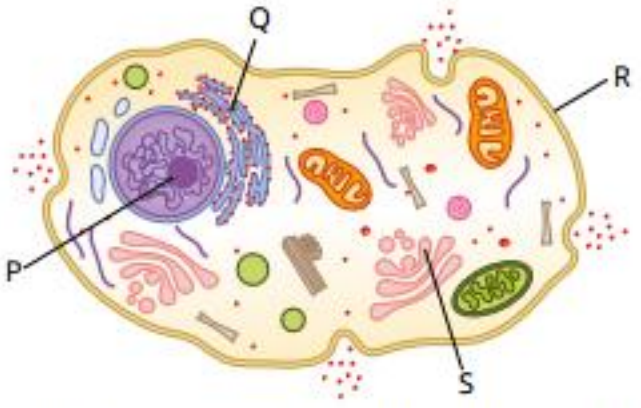
i Identify structures A–I.

ii Identify whether this is a plant cell or an animal cell. Justify your answer.

iii The table below shows a list of functions of organelles. Label the organelles (selected from A–I) with their function.

Organelle	Function
	site of photosynthesis
	stores water in plant cells
	site of lipid production
	contains genetic information and controls the cell's chemical activities
	site of ATP production

b A generalised cell is shown.



i Identify whether this is a prokaryotic or eukaryotic cell. Justify your answer.

ii Identify organelles P–S.

iii Outline how organelles Q–S are involved in the secretion of a protein.

23 **a** An experiment was performed in which muscle cells were incubated in an oxygen-free environment at 20°C. The cumulative uptake of glucose was measured in grams. The results for the first 10 minutes are shown in the following table.

Glucose use in the absence of oxygen

Time (min)	Glucose uptake (g)
2	5
4	10
6	15
8	20
10	25

After 10 minutes, oxygen was infused into the culture and measurement of the uptake of glucose continued. The results for the next 10 minutes are tabulated below. Temperature was maintained at 20°C.

Glucose use in the presence of oxygen

Time (min)	Glucose uptake (g)
12	26
14	27
16	28
18	29
20	30

i Depict on a graph the uptake of glucose versus time for the 20 minutes of the experiment. Clearly mark the point at which oxygen was introduced into the culture.

ii Explain why glucose use declined so significantly after oxygen was added to the culture.

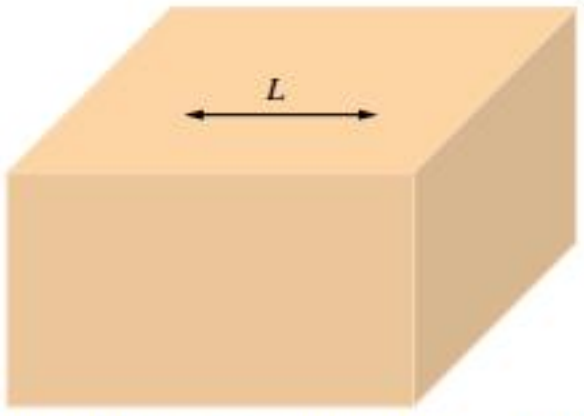
iii Name the independent variable in the experiment.

iv Explain why it was necessary to ensure that the temperature remained at 20°C throughout the experiment.

b Some mitochondrial diseases are caused by mutations in the genes needed for cellular respiration. Mitochondrial diseases caused by these mutations are relatively common in comparison to diseases caused by nuclear chromosomal mutation.

An scientist investigating mitochondrial mutations performed the experiment from part **a** using cells with mitochondria with mutations. The scientist noted that when oxygen was added after 10 minutes to these cells no change in glucose use was observed. Draw a conclusion from these results about the effect of the mutation on mitochondrial function.

24 To investigate the effect of surface area-to-volume ratio on the rate of diffusion, a student prepared different sizes of agar cubes containing phenolphthalein. The agar cubes were then suspended in a 4% sodium hydroxide solution for 10 minutes. When sodium hydroxide diffused into the agar, the agar turned pink. After 10 minutes, the agar cubes were cut in half and the length of the colourless area (L) was measured. The illustration below shows a cross-section of the agar cube.



Surface area of cube = 6 × length of cube × length of cube

Volume of cube = length of cube × length of cube × length of cube

$$\text{Percentage diffusion} = \frac{\text{volume of cube} - (\text{volume of colourless area})}{\text{volume of cube}} \times 100$$

a Calculate and compare the volume, surface area, surface area-to-volume ratio, and percentage diffusion of each cube in the table on the next page.

b Create a graph of percentage diffusion against surface area-to-volume ratio. Deduce and include a curve of best fit on the graph.

c Describe the relationship between surface area-to-volume ratio and diffusion in an agar cube. Use the graph you plotted in part **b** to assist you.

d Identify the size of the cube which was the most effective for maximising diffusion.

e Justify your answer to part **d**.

f A large surface area is essential for quicker diffusion into the cell. Explain why there is a limit to the size individual cells can grow. Justify your answer using the results of the experiment.

g Propose how the reliability and validity of this experiment can be improved.

Length of cube side (cm)	Surface area of cube (cm^2)	Volume of cube (cm^3)	Surface area-to-volume ratio of cube	Length of colourless area, L (cm)	Volume of colourless area ($L \times L \times L$) (cm^3)	Percentage diffusion (%)
1.0				0.0		
1.5				0.4		
2.0				0.8		
2.5				1.8		

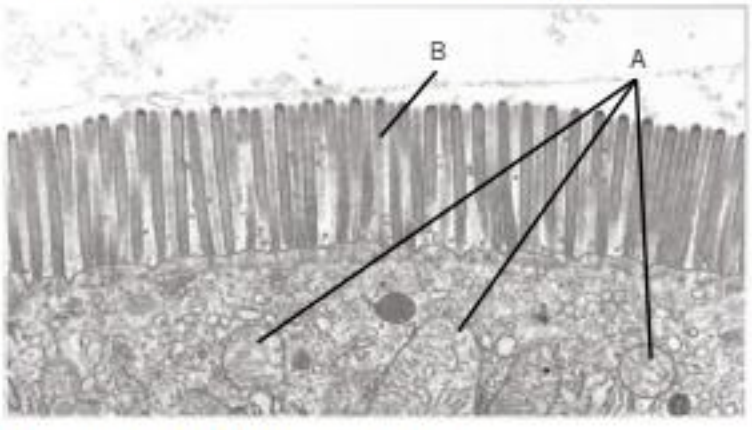
25 A pathologist is examining a blood sample under a microscope and sees four different types of cells. Cell A is 21 µm, cell B is 14 µm, cell C 7.5 µm and cell D 1.0 µm in diameter. Notably cells C and D lack a nucleus. Which cell or cells are prokaryotes? Justify your choice.

26 Many diagrams of animal cells show them as circles, implying that animal cells are mostly spherical, while plant cells are shown as rectangles or squares. A sphere of radius 1.24 mm and a cube of length 2 mm both have a volume of 8 mm^3 rounded to the nearest whole number.

a Use formulas $SA_{sphere} = 4\pi r^2$, $SA_{cube} = 6 \times L^2$, (r = radius of the sphere, L = side length of the cube) to calculate the surface area-to-volume ratio of the two shapes. Use $\pi = 3.14$.

b Use the results of the calculation to explain why such diagrams are misleading.

27 Carefully examine the electron micrograph below.



a Identify the structures labelled A.

b What is the function of structure B?

c How would structures A and B work together to assist the cell to perform its function?

28 **a** State three differences between prokaryotes and eukaryotes.

b Explain the significance of the difference in how the cells function.

29 Consider the substances listed below.

Na^+, CO_2, O_2, C_2H_6O (ethanol), $C_6H_{12}O_6$ (glucose)

a Which substances will cross the cell membrane by simple diffusion?

b Place the substances that cross the membrane by diffusion in order from fastest to slowest movement. Justify your ranking.

c Explain why the other listed substances do not cross by simple diffusion, and state how they enter or leave the cell.

30 Identify the three layers formed during gastrulation and state their ultimate fate.

31 How does the potency of stem cells impact their role in medical interventions to treat damaged tissue?

32 How does cell specialisation contribute to the overall organisation and function of multicellular organisms?

33 Many new parents have the blood from their newborn baby's umbilical cord frozen in liquid nitrogen. Using your knowledge of stem cells, discuss the advantage of retaining umbilical cord blood over blood from an adult.

34 Research is being undertaken into induced pluripotent stem cells. Discuss the advantages of pluripotent stem cells over other types of stem cells, identifying each as an ethical or practical advantage.

Topic 2: Exchange of nutrients and wastes

Multiple-choice questions

1 Select the option that best shows the levels of organisation in a complex organism from simplest to most complex.

A cell → organ → tissue → system
B cell → tissue → organ → system
C system → organ → cell → tissue
D system → tissue → cell → organ

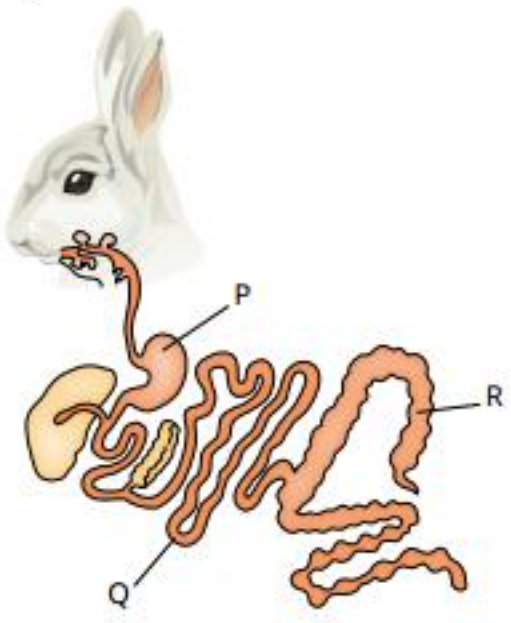
2 Name the part of an organism that contains a large number of different types of cells working together to perform a particular function and that is often seen as a distinct structure.

A tissue
B organ
C system
D organism

3 Select the example of an undifferentiated cell.

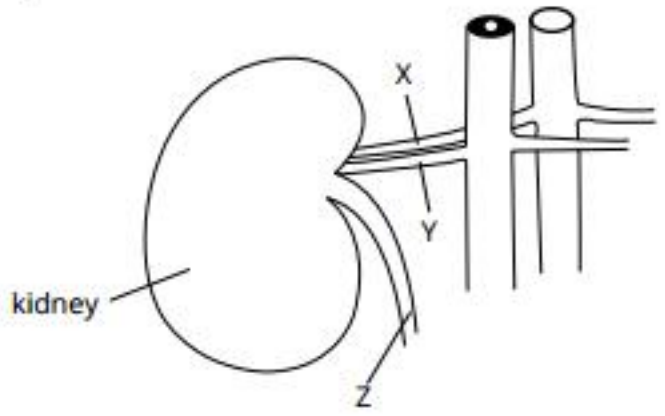
A meristematic cell
B nerve cell
C red blood cell
D mesophyll cell

4 The following diagram shows the digestive system of a rabbit. Identify structures P–R.



	P	Q	R
A	liver	caecum	small intestine
B	stomach	small intestine	caecum
C	liver	small intestine	colon
D	stomach	caecum	large intestine

Questions 5 and 6 refer to the following diagram of a kidney.



5 Identify structures X–Z.

	X	Y	Z
A	artery	vein	ureter
B	artery	vein	urethra
C	vein	artery	urethra
D	vein	artery	ureter

6 Propose which structure in the diagram would contain the highest concentration of urea.

A X **B** Y **C** Z

7 Determine which of the following statements about the changes in the chemical composition of blood as it moves around the body is correct.

A The concentration of glucose increases as it passes through the muscle tissue.
B Fatty acid concentration increases in the blood as it flows through the small intestine.
C The oxygen concentration of the blood decreases as it flows through the brain.
D The bicarbonate ion concentration increases as blood flows through the alveoli.

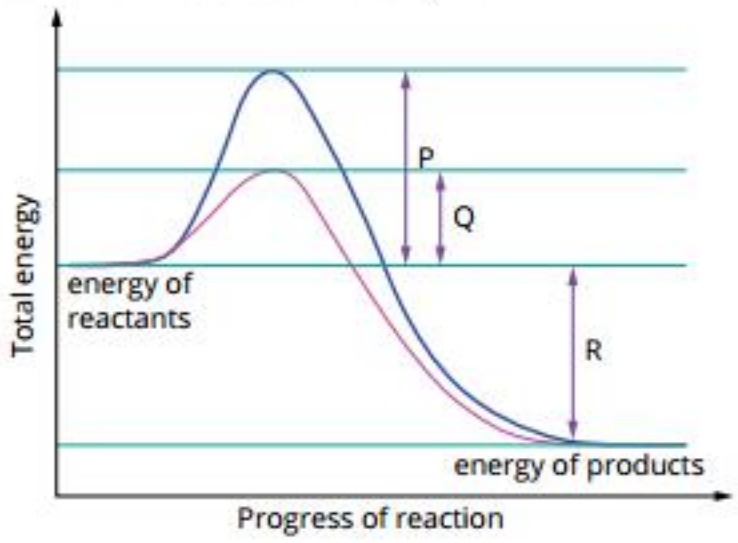
8 Nitrogenous wastes are formed as a result of the breakdown of proteins. The most toxic of these wastes is a substance called creatinine. Name the second most toxic waste.

A urea
B ammonia
C uric acid
D nitric acid

9 Pytalin is a digestive enzyme. It assists in the breakdown of starch to maltose. Name the group of enzymes to which pytalin belongs.

A proteases
B lipases
C amylases
D nucleases

Use the following graph to answer questions 10 and 11. The graph shows the energy levels of a reaction in the presence and absence of an enzyme.



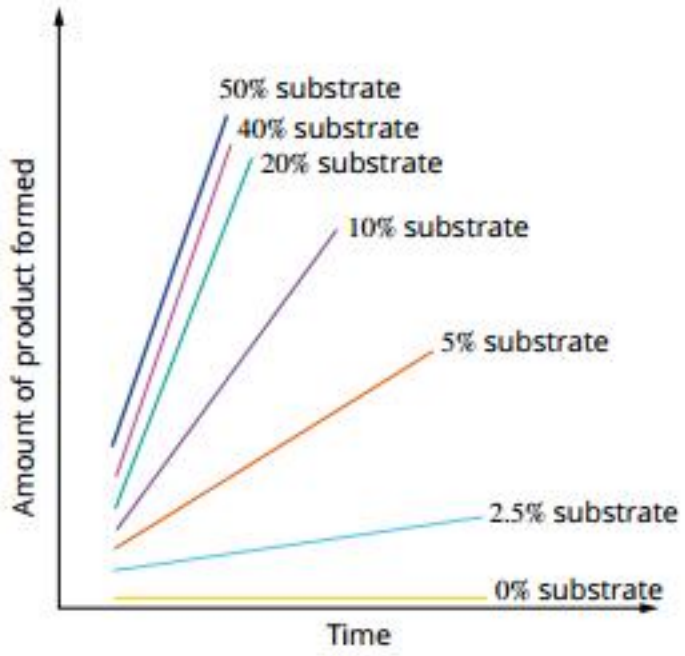
10 Conclude the best explanation for the different energy amounts labelled P, Q and R.

	P	Q	R
A	absence of an enzyme	presence of an enzyme	anabolic reaction
B	presence of an enzyme	absence of an enzyme	catabolic reaction
C	absence of an enzyme	presence of an enzyme	catabolic reaction
D	presence of an enzyme	absence of an enzyme	anabolic reaction

11 Infer which of the following represents the activation energy of the enzyme-catalysed reaction.

A P
B Q
C P + R
D Q + R

12 The following graph shows the effect of changing substrate concentration on the amount of product formed.

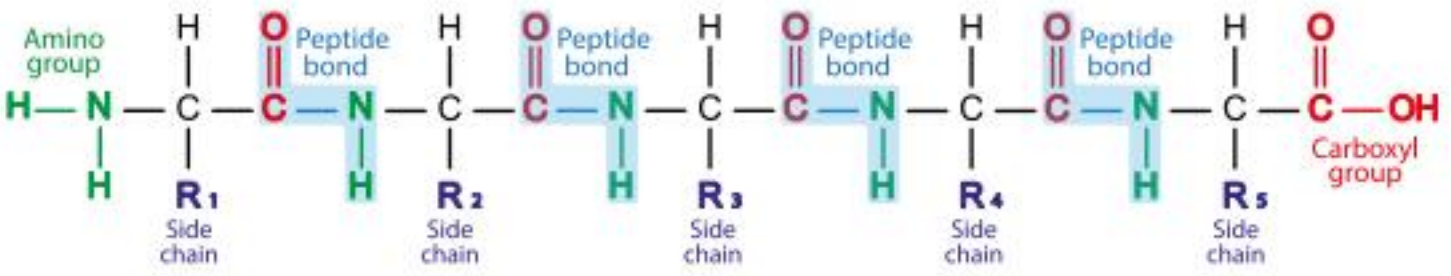


Identify the conclusion that can be drawn.

A The rate of reaction increases exponentially with an increase in substrate concentration.
B The rate of reaction decreases exponentially with an increase in substrate concentration.
C The rate of reaction increases greatly up to a point as the substrate concentration increases, and then the rate of increase starts to decrease.
D The rate of reaction is not affected by any change in the substrate concentration.

13 Identify what kind of organic molecule is shown in the diagram below.

A carbohydrate
B nucleic acid
C lipid
D protein

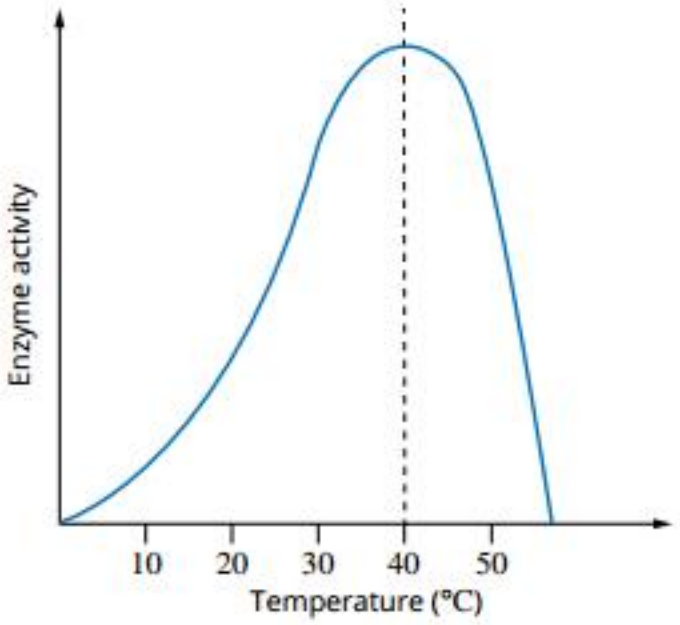


14 State which of the following is a simple carbohydrate.

A fructose
B starch
C chitin
D glycogen

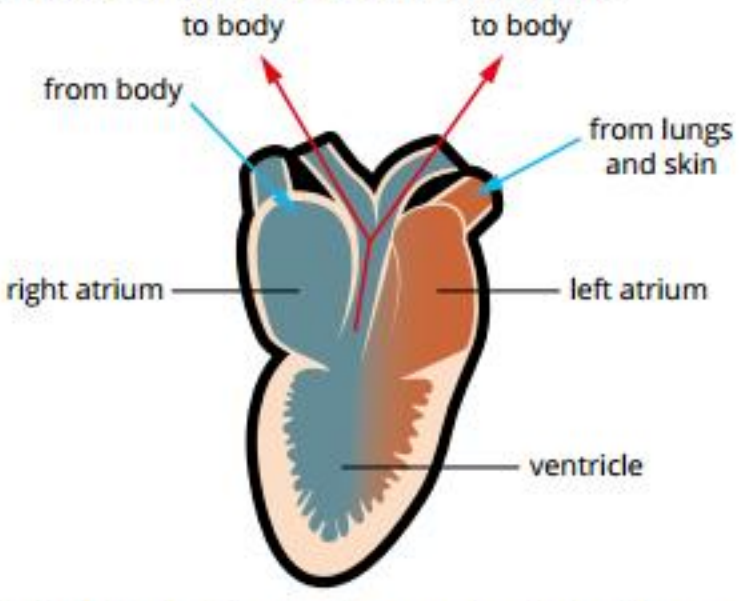
Short-answer questions

15 Study the following graph of enzyme activity.



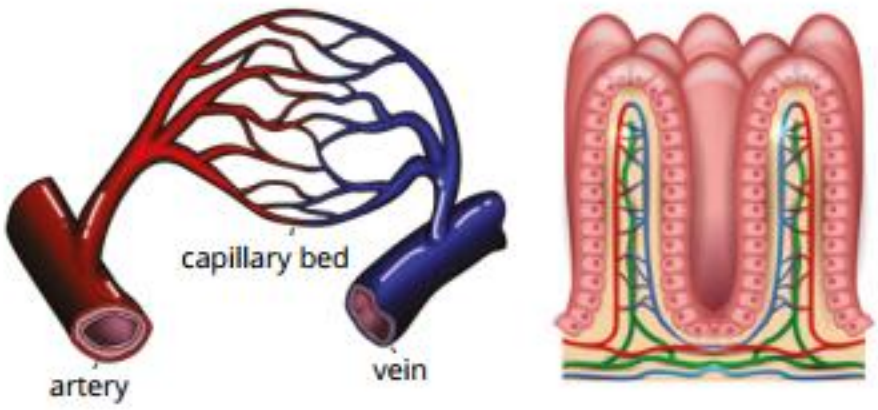
a Outline what happens to enzyme activity as the temperature increases from 0°C to 40°C.
b Identify the optimum temperature for the enzyme.
c Explain what happens to the enzyme above 40°C.
d Name the other factors that affect enzyme activity other than temperature.

16 The following diagram shows the heart of an amphibian. Amphibians have a closed circulatory system.



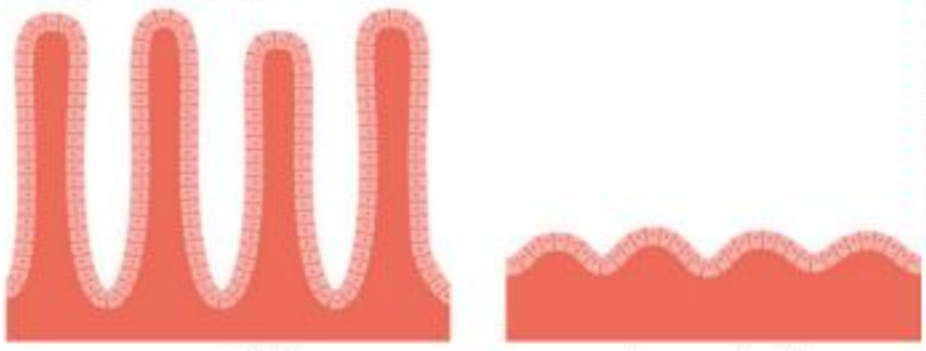
a i Explain what is meant by a closed circulatory system.
ii Identify and explain one advantage of a closed circulatory system.
b i Identify how the amphibian heart differs from the mammalian heart and state how this affects the concentration of gases in the blood going to the tissues.
ii How does this affect gas exchange between the blood and cells in comparison to the exchange in mammals?
c Suggest why a heart structured like that of amphibians would not be suitable for a mammal or bird.

17 Explain what these two structures have in common and relate that commonality to their function.



18 When people with coeliac disease eat gluten, it causes an immune response that damages the villi lining the small intestine. The diagram below shows the difference between normal villi and damaged villi caused by coeliac disease.

Explain the impact of coeliac disease at the:
a cellular level
b tissue level
c organ level
d system level



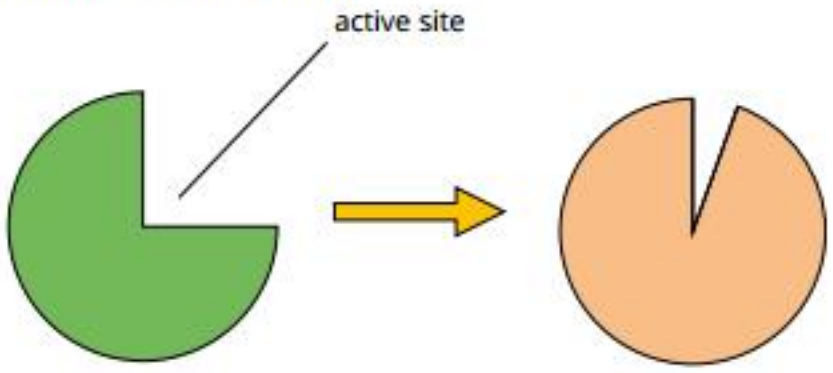
19 The table below shows the changes in composition in fluid as it moves from the blood into the Bowman's capsule and finally into the urine.

Substance	Blood plasma (g/L)	Glomerular filtrate (g/L)	Urine (g/L)
urea	0.3	0.3	12.5
chloride (Cl^-)	3.6	3.6	6.1
sodium (Na^+)	3.2	3.2	6.0
creatinine	0.01	0.01	1.0
protein	72.0	0.0	0.0
glucose	1.0	1.0	0.0
amino acids	0.5	0.5	0.0

Explain what is happening to the various substances and why the changes in concentration are occurring.

20 The blood pressure in a closed system can be maintained at a higher level than in an open system. Suggest how this would affect the flow of materials to and from cells in both systems.

21 An enzyme normally active at pH 4 is exposed to an environment with a pH of 8.6. As a result, the enzyme changed, as shown below.

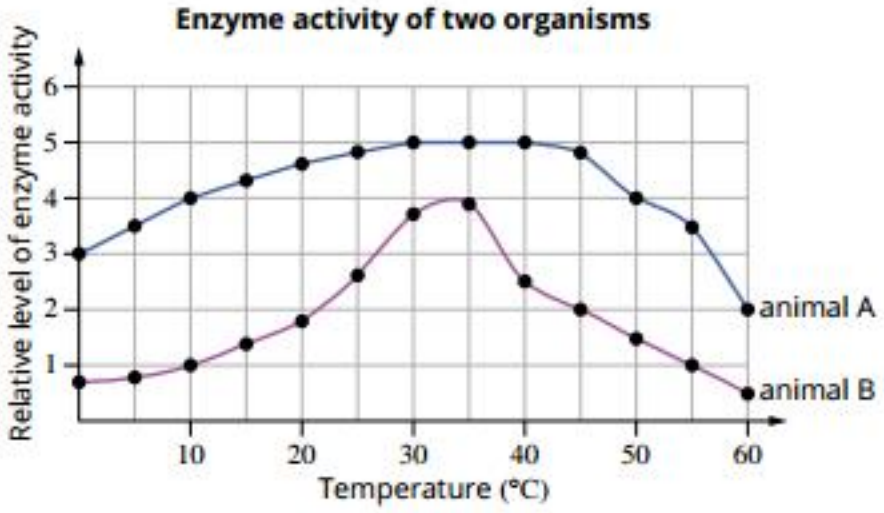


a Explain why the shape changed.

b State how this would affect the activity of the enzyme.

22 The graph below shows the activity of the enzyme catalase in two different animals. Catalase is an intracellular enzyme which breaks down hydrogen peroxide into water and oxygen, according to the equation:

$$2H_2O_2 \rightarrow 2H_2O + O_2$$



a Explain which of the organisms is most likely to be ectothermic.

b Identify the optimum temperature for catalase activity for animal B.

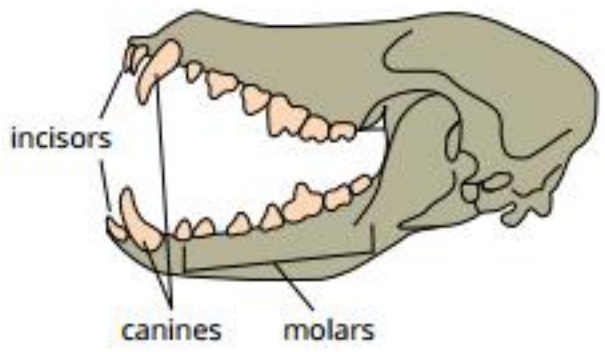
c **i** Explain what is happening to the enzymes of animal B at 55°C.

ii Describe how the relative level of enzyme activity is different at 0°C. Explain why.

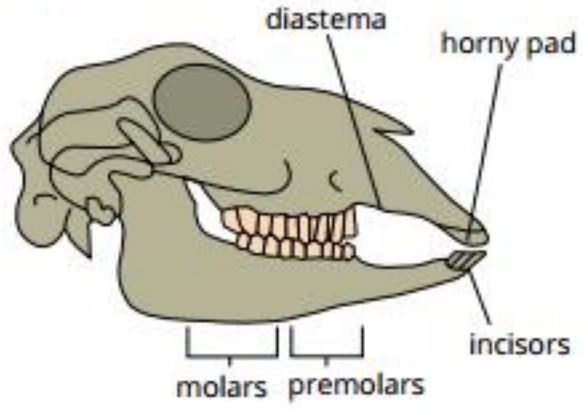
d The graph shows the relative level of enzyme activity. Propose how the experiment could be modified to provide a more exact measure of the activity of catalase.

23 The following diagram shows the skulls of two mammals, organism A and organism B.

Organism A



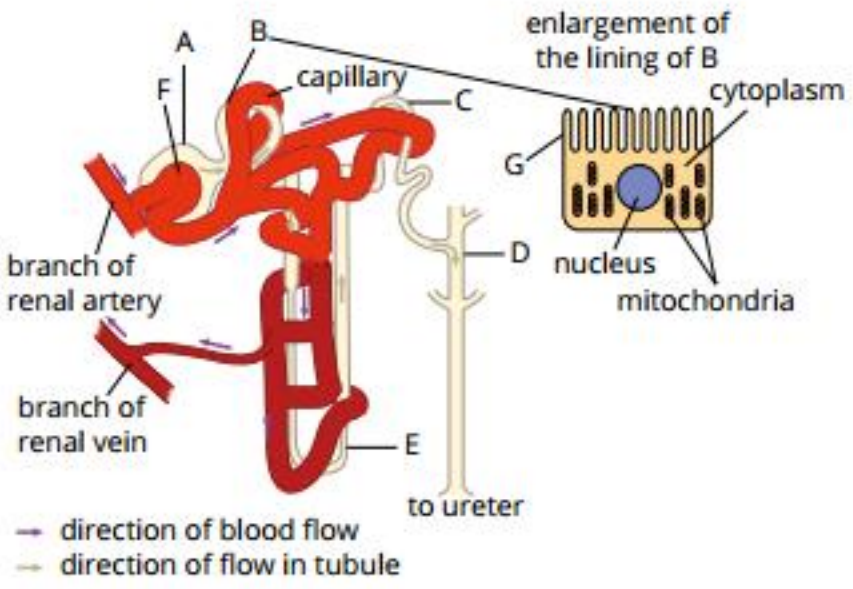
Organism B



a One role of teeth in mammals is to break food into smaller pieces. Explain the benefit of this.

b Describe the type of food that each organism would eat. Justify your opinion and explain the consequences for the structure of their digestive tracts.

24 The kidneys are important in maintaining the osmotic balance of the body and in removing wastes such as the products of protein digestion and excess mineral ions. The functional unit of the kidney is illustrated here.



a Identify structures A–G.

b Explain how structure B assists the nephron to perform its function.

By filtration in the kidneys, 100 litres of fluid is removed from the blood in 24 hours. In this time, only 1.5 litres of urine is formed. The blood plasma contains on average 0.03% urea in solution, but urine contains 2.0% urea in solution.

c Calculate how many times (to the nearest whole number) urea is more concentrated in urine than it is in the blood.

d The table below shows the ratio of nephron concentration to plasma concentration.

Chemical	A	Mid B	Start E	Mid E	End E	Mid C	D
glucose	0.8	0.08	0	0	0	0	0
amino acids	0.4	0.5	0	0	0	0	0
urea	1.0	1.2	1.6	7.0	15.0	20.0	50.0
Cl^-	1.0	1.0	1.0	2.0	0.35	0.4	0.8
Na^+	1.0	1.0	1.0	2.0	0.2	0.25	0.7
K^+	1.0	1.0	1.0	2.0	0.3	0.5	3.0
creatinine	1.0	2.0	5.0	6.5	16.0	20.1	50.0

i Explain what a ratio of 1 for nephron/plasma concentration indicates.

ii Explain why less glucose and amino acids are found in the filtrate than ions such as Na^+ and K^+.

iii Discuss the function of the nephron and its surrounding capillaries. Justify your answer with reference to the data in the table. In your answer, identify how the molecules are being transported across the membrane.

iv Compare the glucose concentration between the renal artery and the renal vein. Justify your proposition.

25 **a** Explain how the structural features of an enzyme enable it to carry out its function.

b The enzyme amylase is present in human saliva and acts on starch as its substrate.

i Which of the three main carbohydrate groups does starch belong to?

ii Deduce why it is important for digestion of starch to begin in the mouth.

c The activity of any enzyme can be affected by factors in its immediate environment.

i Predict what would happen to the amylase activity when pineapple (pH 3.5) and tomato (pH 4) are eaten on a thick-crust pizza that is high in starch content.

ii Design an experiment to investigate how temperature affects amylase activity.

26 A yeast cell is transported from an environment with oxygen to a completely oxygen-free environment.

a Explain the metabolic changes that will occur in the yeast cell.

b Compare the changes in the yeast cell with an animal cell under oxygen-free conditions. Use a diagram to support your answer.

c Predict where and when each of a and b might occur under natural circumstances.

27 Blood and tissue fluid in humans are generally maintained in a narrow pH range, between 7.35–7.45. This means blood is slightly basic.

A family of enzymes called carbonic anhydrases are responsible for the conversion of CO_2 to HCO_3^- in the tissues, and HCO_3^- back to CO_2 in the lungs. Carbonic anhydrase II is responsible for the conversion of CO_2 to HCO_3^-.

Under normal circumstances, carbonic anhydrase can catalyse the change of CO_2 to HCO_3^- at a rate of one million molecules per second. Imidazole is a chemical that inhibits the action of carbonic anhydrase II. An experiment was run at 37°C, with and without imidazole, at varying CO_2 concentrations. The amount of imidazole added was kept constant throughout the experiment. The results are shown in the table below.

CO_2 concentration ($mmol\,L^{-1}$)	Rate of conversion	
	Without imidazole ($mmol\,min^{-1}$)	With imidazole ($mmol\,min^{-1}$)
0.1	3.2	1.6
0.2	4.5	2.7
0.3	6.7	4.1
0.5	7.1	6.3
1.0	8.8	7.8
2.0	10.3	10.1

a i Name the independent variable(s) in this experiment.

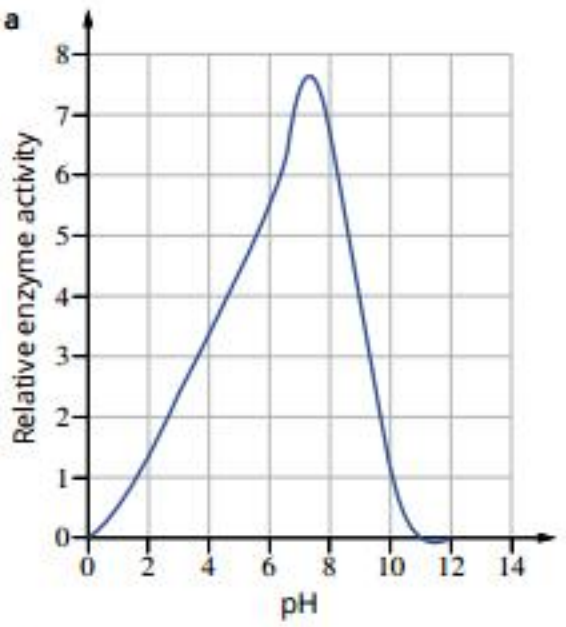
ii Imidazole is a competitive inhibitor of carbon dioxide conversion. Explain how imidazole slows the rate of reaction. Ensure that you describe the structures involved in your answer.

iii Explain the difference in the rate of inhibition as the concentration of carbon dioxide changes.

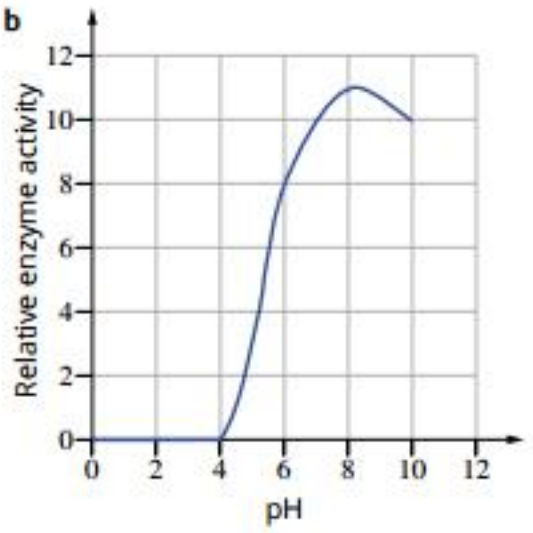
b In a further experiment on the activity of carbonic anhydrase II, its activity during changes to pH was investigated. The results were graphed. Four graphs are shown below.

Propose which of the graphs is most likely to show the activity of this enzyme at various pH values. Justify your choice and explain what is occurring to the enzyme. State why the other graphs are not correct.

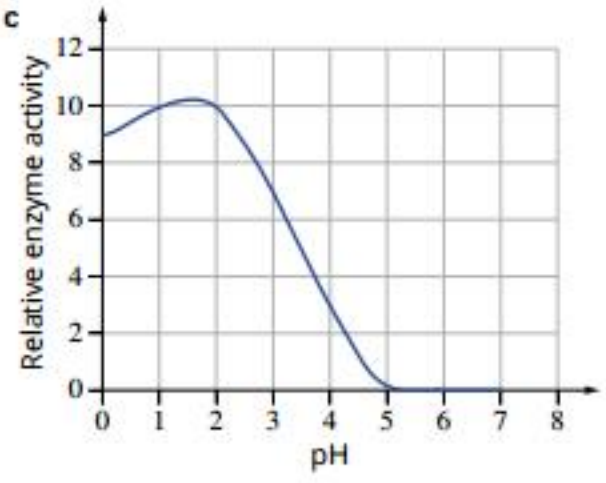
a



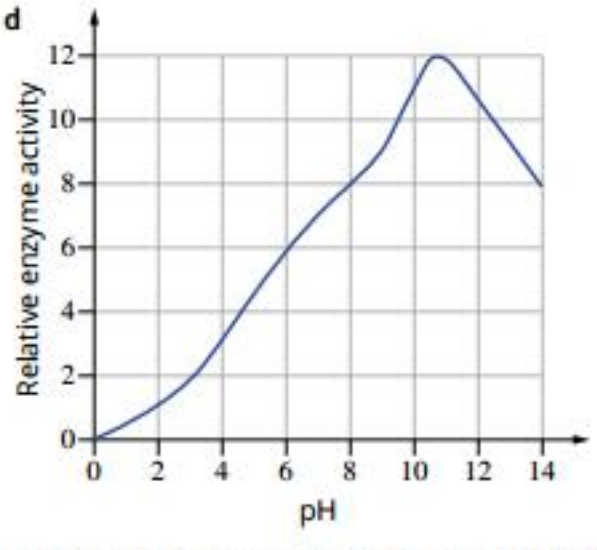
b



c



d



c Imidazole could be used as a poison. Explain how it would affect the metabolism of an organism.

Topic 3: Cellular energy, gas exchange and plant physiology

Multiple-choice questions

1 Identify which of the following is not a product of the Krebs cycle.
- **A** $NADPH + H^+$
- **B** $NADH + H^+$
- **C** CO_2
- **D** ATP

2 Which of the following situations is anabolic in nature?
- **A** anaerobic respiration
- **B** the breakdown of lipids during digestion
- **C** Calvin cycle
- **D** Kreb's cycle

3 Which of the following is *not* a product of anaerobic respiration?
- **A** CO_2
- **B** lactic acid
- **C** alcohol
- **D** water

4 During cellular respiration most ATP is produced during:
- **A** anaerobic respiration
- **B** the electron transfer chain
- **C** Kreb's cycle
- **D** glycolysis

5 Carbon monoxide (CO) is toxic to cells as it has the ability to bind firmly to cytochrome c, stopping its activity. An experiment was undertaken in which plant cells in tissue culture, without light, were exposed to high levels of CO. Which of the following results would be expected?

	Oxygen usage	Ethanol acid concentration	CO_2 concentration
A	increased	increased	increased
B	decreased	unchanged	increased
C	increased	increased	unchanged
D	decreased	increased	increased

6 Which of the following are produced during the Calvin cycle?
- **A** GAP, ADP, $NADP^+$
- **B** glucose, $NADP^+$, CO_2
- **C** ATP, NADPH, O_2
- **D** $NADP^+$, ATP, O_2

7 NADH and ATP are both produced in
- **A** the cristae of mitochondria
- **B** the matrix of mitochondria
- **C** the grana of chloroplasts
- **D** the stroma of chloroplasts

8 Which of the following statements about photosynthesis is *not* true?
- **A** ATP is made during the light reactions.
- **B** The oxygen released during photosynthesis is produced when water is split.
- **C** The light reactions can only happen in the light and the dark reactions can only happen in the dark.
- **D** ATP is an input into the Calvin cycle.

9 Which of the following equations represents the overall reactions of aerobic respiration?
- **A** $C_6H_{12}O_6 \rightarrow$ alcohol + 2 CO_2 + 2 ATP
- **B** $C_6H_{12}O_6 \rightarrow$ lactic acid + 2 ATP
- **C** $C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O$ + 36/38 ATP
- **D** $6CO_2 + 6H_2O \rightarrow C_6H_{12}O_6$ + 36/38 ATP

Short-answer questions

10 An experiment was undertaken to investigate photosynthesis and cellular respiration using algal balls. Half teaspoons of algal balls were placed in two sealed containers (tubes) of soda water and indicator solution. Soda water contains dissolved carbon dioxide. One tube was placed in bright light and the other was placed in the dark. A third tube containing only soda water and indicator (and no algal balls) was placed in the light. When carbon dioxide dissolves in water, it forms carbonic acid. Acids have a low pH. The colour of the solution was matched to a colour chart to determine the pH throughout the experiment. The results of the experiment are shown in the table below.

Time (minutes)	0	30	60	90	120	150	180	210
pH with algal balls in bright light	8.2	8.4	8.5	8.7	8.9	9.0	9.0	9.1
pH with algal balls in the dark	8.2	8.3	8.2	8.0	7.8	7.7	7.6	7.6
pH without algal balls	8.2	8.2	8.2	8.2	8.2	8.2	8.2	8.2

a Why was a tube without algal balls used?

b Explain why the pH in the tube in the light is rising.

c The experiment was run for 3.5 hours. If the experiment had continued for a further 24 hours, would you expect the results for the tube with algal balls in the light to be any different? Explain.

d **i** Suggest a possible reason why the pH in the tube in the dark rose after 30 minutes.

ii Why might the pH have stopped changing in the tube in the dark after 180 minutes?

e How might this experiment be improved?

11 Complete the following table showing where ATP is made in a cell and its fate.

Site	Y if made there, N if not	Fate of any ATP produced
cytosol		
Kreb's cycle		
electron transfer chain of mitochondrion		
thylakoids		
stroma		

12 **a** Guard cells form the stomata of cells. They have unique features that enable them to perform their function. State what these features are and describe how they enable stomata to regulate gas exchange.

b Researchers investigating the effects of rising CO_2 levels on stomatal density surveyed 100 different species and found an average reduction in stomatal density of 14.3%. Using your knowledge of the role of stomata in gas exchange, explain why this reduction may have occurred.

13 A mitochondrion is described as having folded inner membranes that increase surface area.

a Explain why this is an advantage to a cell.

b Does a chloroplast in a plant cell also have structures that increase the surface area for its function?

c Draw labelled diagrams to support your answers to **a** and **b** above.

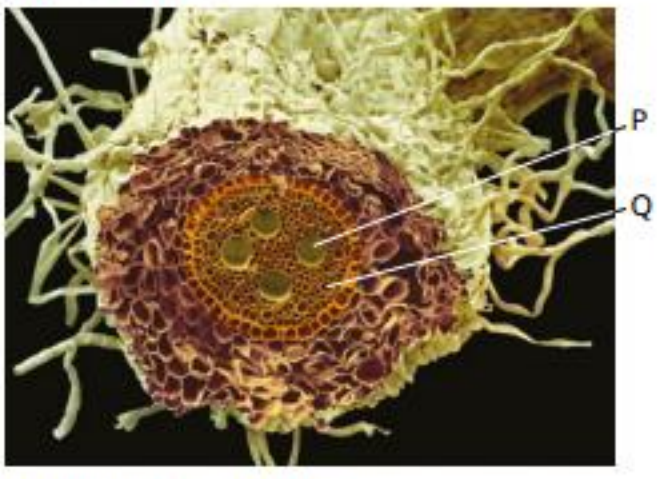
14 Compare anaerobic and aerobic cellular respiration.

15 List the features of an efficient respiratory membrane and explain how they assist gas exchange.

16 **a** Identify the three main components of an ATP molecule.

b Explain the process in which ATP releases energy for chemical reactions.

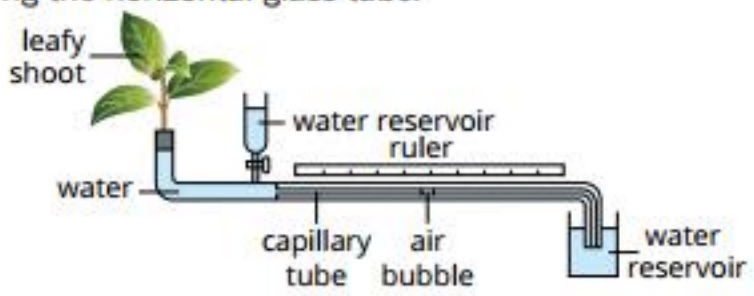
17 The following image shows a scanning electron micrograph of a root.



a Identify structures P and Q.

b Outline how structure P is involved in the transport and eventual loss of water.

18 The diagram shows an apparatus called a plant potometer. It is used to measure the rate of transpiration of a plant by timing the rate at which the bubble moves along the horizontal glass tube.

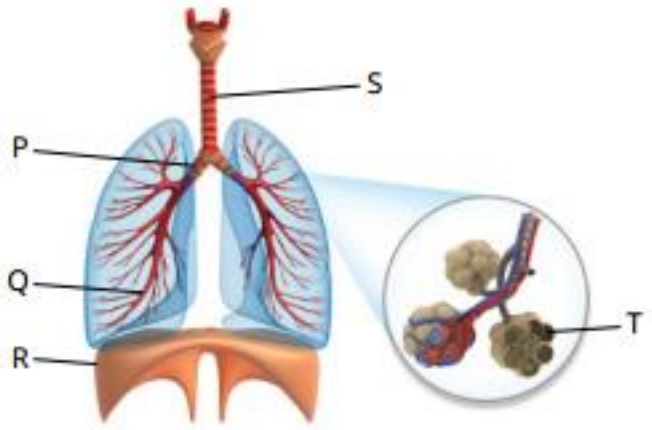


a Design a controlled experiment using a plant potometer to test the hypothesis that the leaves of Australian eucalypts do not lose water as fast as leaves of an English elm. In your answer mention the test and controlled variables.

b Propose the results you would expect if the hypothesis were to be supported.

19 Compare xylem and phloem.

20 **a** The following diagram shows the structure of the human respiratory system.



i Identify structures P–T.

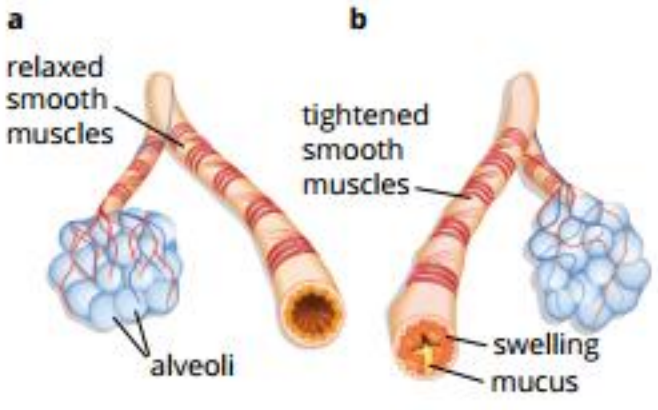
ii Describe how the movement of the rib cage and diaphragm results in air moving into T.

iii Describe the features of T that make it a suitable surface for the exchange of gases.

b **i** Draw a diagram showing how gas is exchanged between the capillary net surrounding the air sac and the interior of the air sac.

ii Compare the flow of gases at the body tissues, such as muscles, and gas flow in the lungs.

c This diagram shows structure P during normal breathing (A) and during an asthma attack (B).

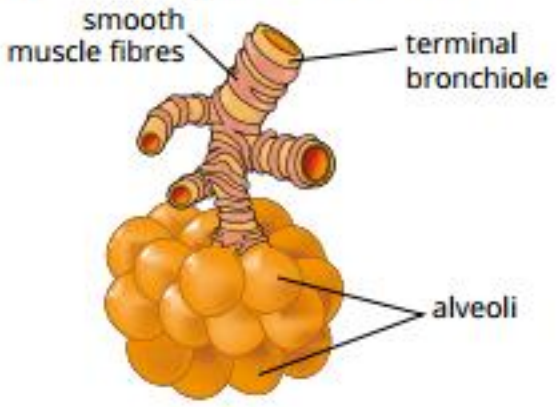


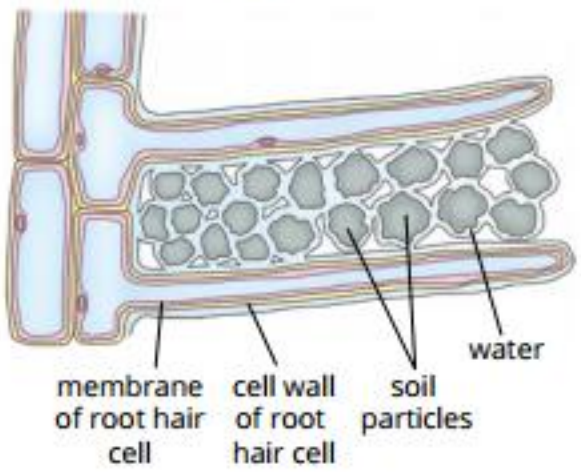
i List the conditions in a person's alveoli during an asthma attack.

ii Explain the reason for the conditions listed in part **c (i)**.

iii Asthma interferes with one important component of the gas exchange process. Discuss.

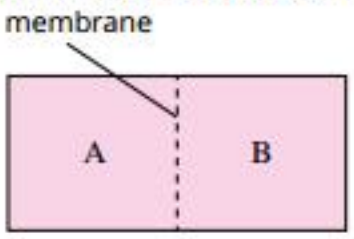
21 Explain what these two structures have in common and relate that commonality to their function.





22 a A pressurised container has a total pressure of 980 mmHg. The container contains 25% oxygen, 40% nitrogen and 35% carbon dioxide. Calculate the partial pressure of each gas.

b A second container is joined to the first, as shown.

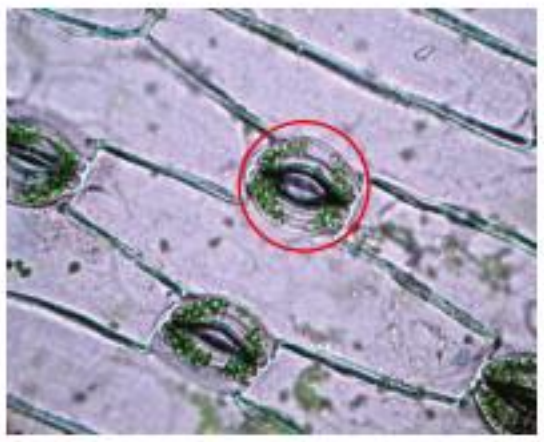


The two containers have a membrane between them that is permeable to all of the gases. The pressure in the second container is 600 mmHg. This container contains 60% oxygen and 40% nitrogen.

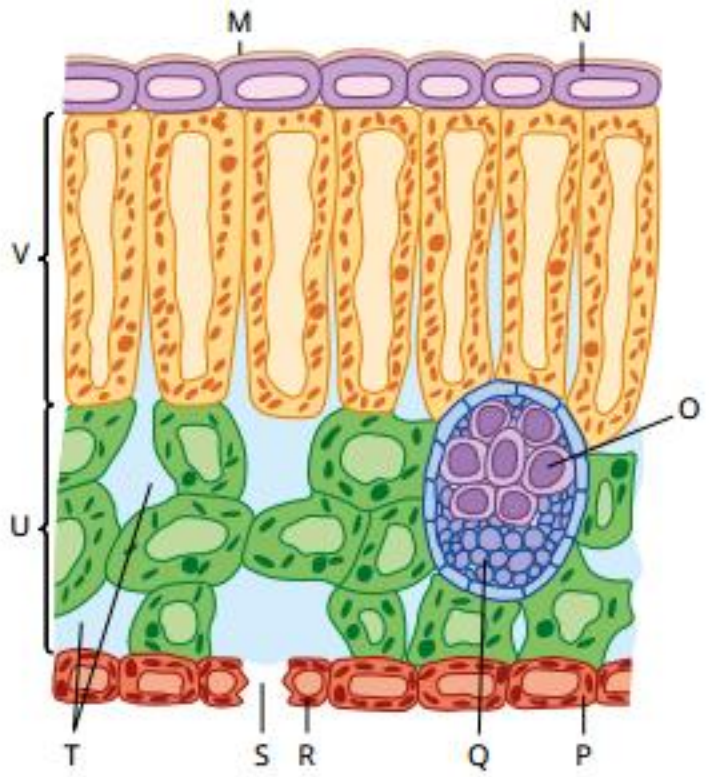
i Calculate the partial pressures of both gases in the container.

ii Conclude which way each of the gases will move.

23 a The image below is a micrograph of a leaf epidermis. Describe the function of the circled structure.

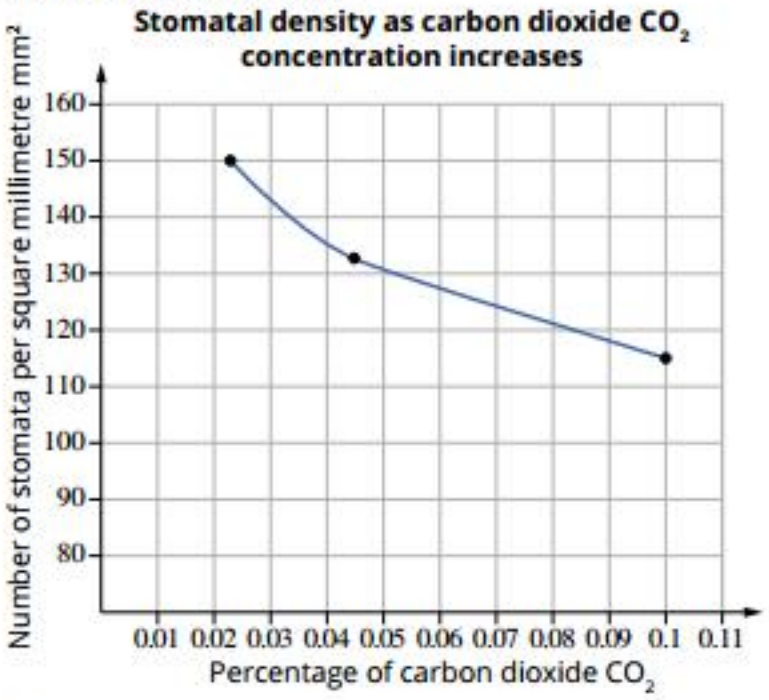


b Identify structures M–V.



c Explain the disadvantage plant of structure S.

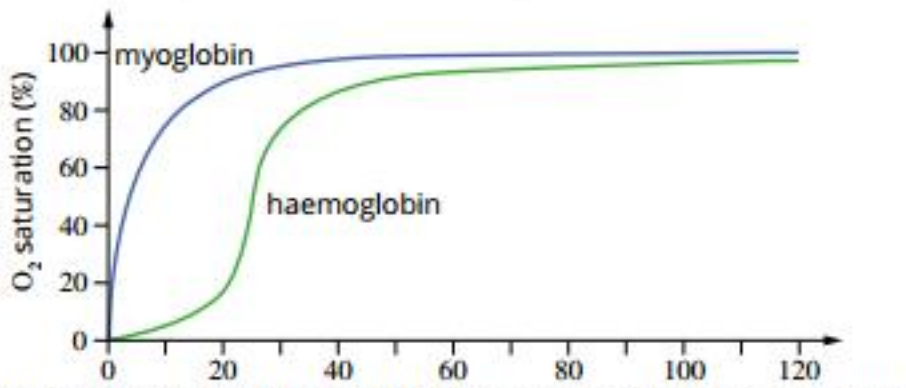
d Analyse the graph below.



i Describe the trend in the number of stomata as carbon dioxide concentration increases.

ii Propose what advantage exists for the plant that results in this trend.

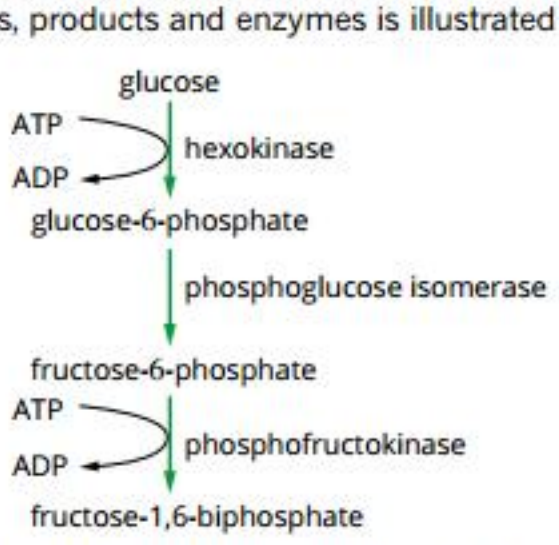
24 Haemoglobin is the oxygen-carrying pigment in red blood cells that gives them their red colour. Myoglobin is an oxygen-carrying pigment found in muscle tissue. It is found in particularly high concentrations in the muscles of diving mammals such as dolphins and whales.



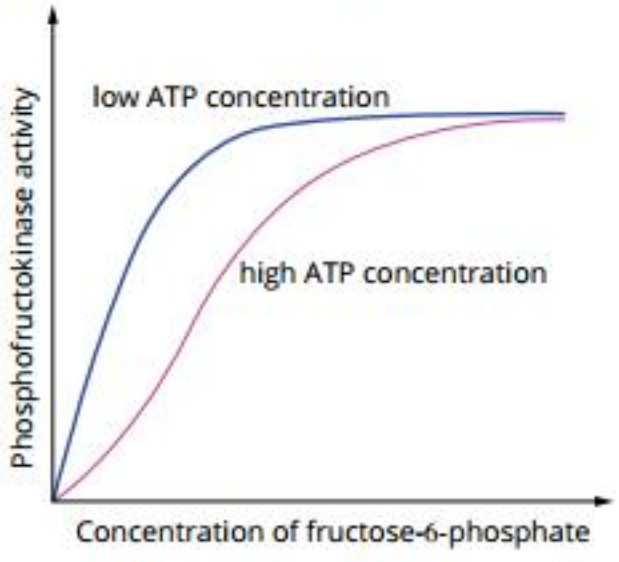
a Explain why haemoglobin is well suited to its role of collecting oxygen in the lungs and releasing it in the tissues. Refer to the data in your answer.

b Diving mammals have large amounts of myoglobin in their muscles. Propose how this helps them to stay underwater for long periods of time. Justify your answer using the data supplied.

25 A section of the glycolysis pathway showing various substrates, products and enzymes is illustrated here.



Scientists measured the effect of different concentrations of fructose-6-phosphate on phosphofructokinase activity. Phosphofructokinase activity was also measured with low and high concentrations of ATP in the reaction mixture. The graph below shows the results.



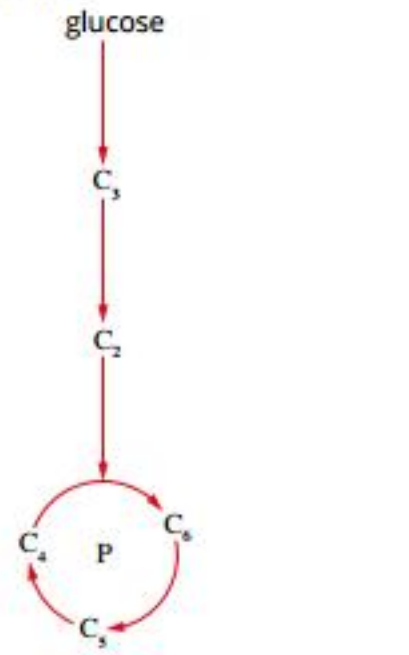
a Outline the effect of increasing fructose-6-phosphate.

b Explain how increasing the concentration of fructose-6-phosphate affects the activity of phosphofructokinase.

c Outline the effect of increasing ATP concentration on the activity of phosphofructokinase.

d Given that one of the functions of glycolysis is to produce ATP, explain how the effect of ATP on phosphofructokinase is advantageous.

26 The following diagram shows part of the cellular respiration pathway. The number of carbon atoms in each molecule is indicated.

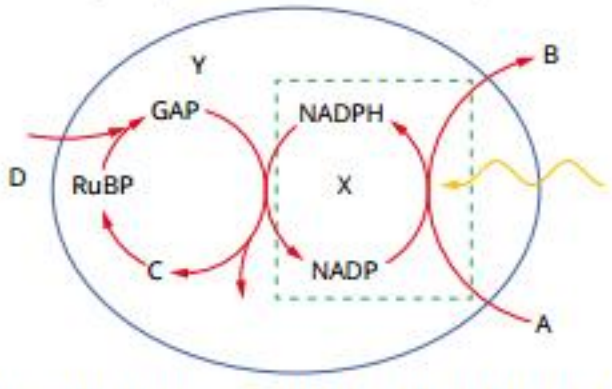


a Label pyruvate and acetyl CoA on the diagram.

b Circle the part of the process that occurs in the matrix of the mitochondrion on the diagram.

c Identify process P.

27 The following diagram shows an outline of a chloroplast, with the letters A—D representing the chemicals involved in photosynthesis. Different compartments are represented by X (within the dotted line) and Y.



a Identify substances A to D from the following list: CO_2, O_2, H_2O, glucose (G3P)

b i Identify compartments X and Y.

ii Name the group of reactions that happen in each compartment.

c Propose which of these substances would be in short supply on an overcast day. Justify your proposal.

d Suggest how the production of glucose would be affected.

28 A group of students interested in plant transport systems performed two radioactive tracing experiments. Radioactive isotopes of elements act exactly the same as the non-radioactive forms but are able to be traced through the plant.

In the first experiment, the students grew the plants in a medium containing nitrates labelled with a radioactive isotope of nitrogen. In the second experiment, they grew the plants in an environment with an atmosphere containing carbon dioxide labelled with carbon-14, which is radioactive.

Experiment 1

All of the plants were grown in a full nutrient solution containing radioactively labelled nitrates. The nutrient medium of half of the plants contained a low concentration of arsenic. Arsenic is a poison that disrupts the electron transfer chain of cellular respiration. The reduction in concentration of nitrates in the nutrient medium was monitored and registered every 10 minutes.

a **i** Identify the control in this experiment.

ii Name the independent variable.

b The results of the experiment are shown in the table.

Time (min)	Nitrate concentration of nutrient medium for 'No arsenic' plants ($mmol\,L^{-1}$)	Nitrate concentration of nutrient medium for arsenic-treated plants ($mmol\,L^{-1}$)
0	1.1	1.1
10	1.0	1.05
20	0.85	1.0
30	0.8	0.98
40	0.75	0.97
50	0.6	0.97
60	0.5	0.97

i Draw a graph of the results.

ii Explain where you would first expect to see radioactive nitrogen in the plants.

iii Draw conclusions about nitrate absorption into plants from this experiment.

iv After 48 hours, the plants without arsenic were examined to discover the distribution of radioactive nitrogen in their tissues. Describe where in the plant would you expect to see evidence of radioactivity. Justify your suggestion.

Experiment 2

Mangrove plants use structures called pneumatophores to allow roots to undergo gas exchange. The canopy of a small mangrove plant was covered in a tent that had a constant supply of air. The carbon dioxide in the air contained radioactive ^{14}C rather than the normal ^{12}C. The pneumatophores were covered separately to collect any gases that were released by them.

d Explain why the tents over the pneumatophores were filled with oxygen.

e Several hours after the start of the experiment the tents over the pneumatophores showed evidence of radioactivity.

i Name the compound that contained the radioactive material.

ii Explain in detail the pathway taken by the ^{14}C to get to the tent over the pneumatophores.

29 Sea stars have two stomachs. Part of the cardiac stomach can be pushed out of its mouth to grab prey or be inserted between shells to obtain what is inside of the shells. When the material is inside the cardiac stomach, strong digestive enzymes begin breaking it down. Once the cardiac stomach is back inside the sea star, its contents are pushed into the pyloric stomach to be further digested.

a Determine which stomach would be better having a thicker external wall. Justify your reasoning.

b Determine which stomach would be better having a high surface area-to-volume ratio. Justify your reasoning.

c Sea stars do not have true lungs or true gills. Hypothesise how oxygen might get into the sea star's body from the surrounding water.

30 A teacher moved from Victoria to Queensland. She had grown some excellent peace lilies, *Spathiphyllum sp.* in Victoria, and she began growing them in Queensland when she moved there in December. She knew they did not like much water so was careful to water them only once a week, giving them enough water to make their soil damp. After 3 weeks, they did not look very well, so she began to water them twice a week. However, the plants soon became droopy and looked very unwell. The teacher asked the local plant nursery for advice.

a Comment on the weather in Queensland during December and January.

b Propose how the transpiration stream would be affected by the weather during this time.

c Hypothesise what might be happening in the pot and soil where these plants were growing.

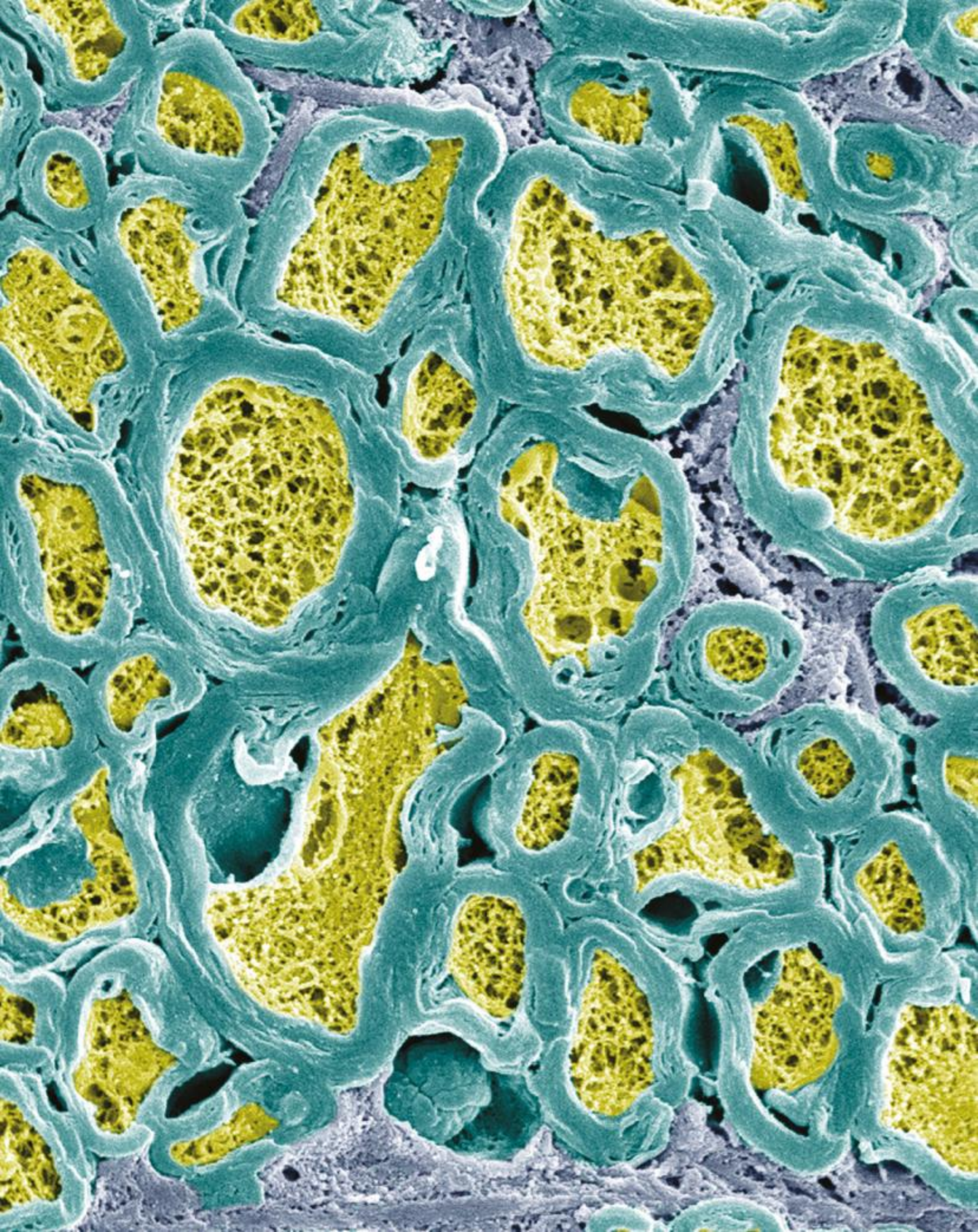
d Predict what advice the nursery might provide.

31 A teacher was demonstrating the inflation of cow lungs to students in the Year 11 Biology class. They attached one end of a tube to a pump and placed the other end through the trachea and down into the bronchus. The teacher then began to pump.

a Predict what happened next.

b One student felt that what they saw was a trick and that the teacher was actually using something else to push up the lung. Propose a reason why the student was amazed and dubious about what they saw.

c Identify another feature of efficient gas exchange that has been demonstrated by this activity.



UNIT 2 Maintaining the internal environment

TOPIC 1 Homeostasis—thermoregulation and osmoregulation

TOPIC 2 Infectious disease and epidemiology

Unit 2 objectives

- Describe ideas and findings about homeostasis, thermoregulation and osmoregulation, infectious disease and epidemiology.
- Apply understanding of homeostasis, thermoregulation and osmoregulation, infectious disease and epidemiology.
- Analyse data about homeostasis, thermoregulation and osmoregulation, infectious disease and transmission and epidemiology.
- Interpret evidence about homeostasis, thermoregulation and osmoregulation, infectious disease and epidemiology.
- Evaluate processes, claims and conclusions about homeostasis, thermoregulation and osmoregulation, infectious disease and epidemiology.
- Investigate phenomena associated with homeostasis, thermoregulation and osmoregulation, infectious disease and epidemiology.

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CHAPTER

06 Homeostasis

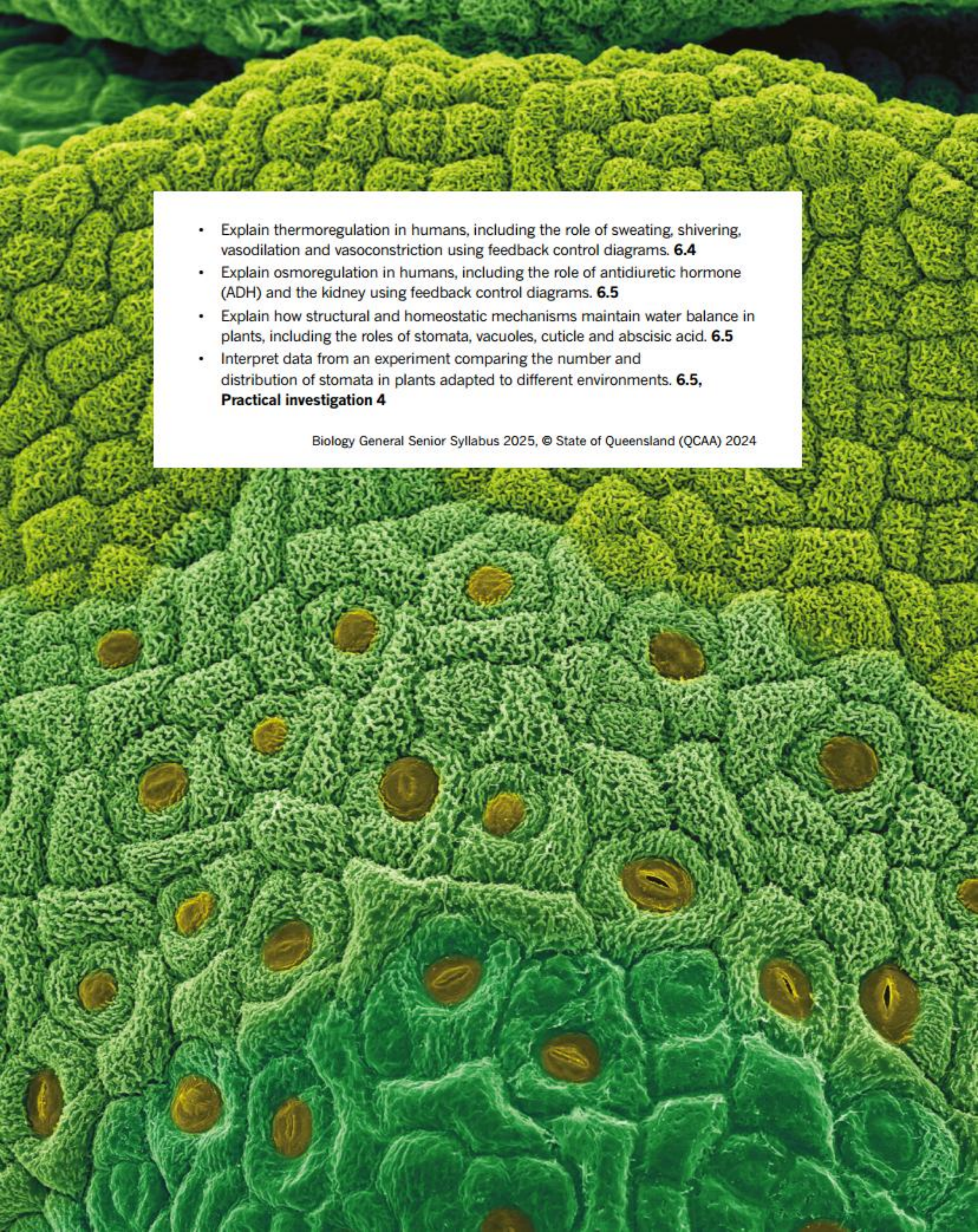
Organisms and cells are constantly experiencing changes in their environment. These changes to the internal and external conditions can adversely affect the survival, growth and function of the organism. The internal environment of an organism must always remain within tolerable limits, even when conditions in the external environment fluctuate widely. This is the ability to maintain homeostasis.

Living organisms rely on their external environments to provide adequate levels of nutrients, water and oxygen, and suitable physical conditions, such as light and temperature. Organisms have a range of mechanisms that allow them to adapt to changing conditions while maintaining a stable internal environment when external conditions fluctuate. Both the nervous system and endocrine systems of an organism can act to maintain homeostasis. If an organism is not able to adapt to its external environment, it will suffer cellular damage and possibly death when conditions change.

Syllabus subject matter

Topic 1 • Homeostasis—thermoregulation and osmoregulation

- Explain how the nervous and endocrine systems use negative feedback to coordinate responses to internal/external stimuli and maintain homeostasis (stimulus–response model). **6.1**
- Identify the different types of sensory receptors and their stimuli, including chemoreceptors, thermoreceptors, mechanoreceptors, photoreceptors and nociceptors. **6.1**
- Describe the structure and function of nerve cells, including dendrites, soma, body, axon, myelin sheath, nodes of Ranvier, axon terminal and synapse. **6.2**
- Distinguish between sensory neurons, interneurons and motor neurons. **6.2**
- Explain the passage of a nerve impulse in terms of transmission of an action potential and synaptic transmission, referring to neurotransmitters, receptors, synaptic cleft, vesicles, postsynaptic and presynaptic neurons and signal transduction. **6.2**
- Describe how hormones relay messages to cells displaying specific receptors via the circulatory or lymphatic system. **6.3**
- Explain how receptor binding alters cellular activity, recognising that a cell's sensitivity to a specific hormone is directly related to the number of receptors it displays for that hormone. **6.3**
- Analyse feedback-control diagrams to identify the stimulus, receptor/s, control centre, effectors and communication pathways in different scenarios. **6.1, 6.2, 6.3, 6.4, 6.5**
- Explain thermoregulatory mechanisms of endotherms, including:
  - structural features: brown adipose tissue, insulation
  - behavioural responses: kleptothermy, hibernation, aestivation and torpor
  - physiological mechanisms: evaporative heat loss, thermogenesis and vasomotor control. **6.4**



- Explain thermoregulation in humans, including the role of sweating, shivering, vasodilation and vasoconstriction using feedback control diagrams. **6.4**
- Explain osmoregulation in humans, including the role of antidiuretic hormone (ADH) and the kidney using feedback control diagrams. **6.5**
- Explain how structural and homeostatic mechanisms maintain water balance in plants, including the roles of stomata, vacuoles, cuticle and abscisic acid. **6.5**
- Interpret data from an experiment comparing the number and distribution of stomata in plants adapted to different environments. **6.5, Practical investigation 4**

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6.1 Maintaining homeostasis

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain how the nervous and endocrine systems use negative feedback to coordinate responses to internal/external stimuli and maintain homeostasis (stimulus–response model)
- identify the different types of sensory receptors and their stimuli, including chemoreceptors, thermoreceptors, mechanoreceptors, photoreceptors and nociceptors.

Homeostasis is when variables in a system are maintained within certain limits. When an organism is healthy and functioning well, its systems are in homeostasis. But when unwanted changes to these limits occur, homeostasis can be achieved by detection of changes by different receptors and adjustment to changes. The process of homeostasis results in a variety of mechanisms that work together to keep **internal environments** constant. The ability for an organism to achieve balance internally maintains conditions at an optimum level when the internal or **external environment** changes.

Different types of sensory **receptors** can detect changes to different stimuli in the environment. These receptors include **chemoreceptors** (which detect changes to chemical compounds), **thermoreceptors** (which detect change in temperature), **mechanoreceptors** (which detect change in pressure and movement), **photoreceptors** (which detect change in light) and **nociceptors** (which detect pain).

Once stimuli have been detected by receptors, an organism can coordinate a response (via the stimulus–response model) to bring about changes needed to maintain homeostasis. The **nervous system** and **endocrine system** (hormonal system) are both able to play a role in coordinating a response.

The ability to maintain these optimum levels is dictated by the organism's metabolism—the complex integrated network of biochemical reactions that sustain life that are regulated by enzymes. As you will recall from Chapter 3, enzymes work within optimal conditions. Homeostasis includes the maintenance of these conditions for both catabolic and anabolic reactions.

The endocrine system consists of glands, tissues and organs that produce and regulate hormones.

RESPONDING TO CHANGING ENVIRONMENTS

Homeostasis (from the Greek *homoios* and *stasis*, meaning 'staying in the same place') is achieved by negative feedback loops. In **negative feedback loops**, receptors detect a change in the internal environment (**stimulus**), and **effectors** work to reverse the direction of the change to achieve equilibrium. In positive feedback loops, the effectors work to maintain and enhance the direction of the stimulus; therefore, positive feedback is not usually involved in maintaining homeostasis. Complex multicellular animals maintain the equilibrium of their internal environments by detecting and responding appropriately to the many changes in the external and internal environment.

Animals coordinate the activities of their cells, tissues and organ systems so that responses occur in an integrated and controlled manner. Detecting and responding to a stimulus requires an effective internal communication system. Communication in animals is achieved by hormonal and nervous system mechanisms, which transmit information between different parts of the organism, and also translate environmental disturbances into signals that can be interpreted and responded to. For example, the iris of the human eye detects light, **neurons** receive this signal, and the eye responds by dilating or constricting using muscles of the eye to regulate the amount of light that enters the eye (Figure 6.1.1).

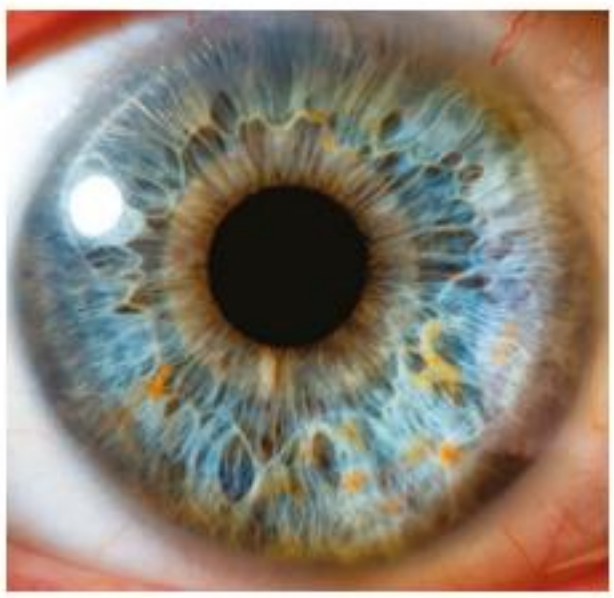


FIGURE 6.1.1 The iris is a pigmented muscle that responds to a light stimulus by dilating and constricting to regulate the amount of light entering the eye.

These regulatory systems are the most developed in mammals, which are able to maintain a relatively stable internal environment in the face of changing conditions.

Although in many ways hormonal systems and nervous systems appear distinctly different, they share one common feature: they both involve chemical communication. In both systems, signals are passed from one cell to the next by the release of specific molecules, known as hormones and neurotransmitters. Hormones are released from glands or other tissues, and neurotransmitters are released from nerve endings. These molecules exert their effects by highly specific interactions with a receptor found on, or within, the responding or target cell.

Feedback loops

Feedback loops may involve the endocrine and nervous systems working together to regulate the internal environment.

Negative feedback loops promote stability in the internal environment and maintain homeostasis by responding to changes in the body and adjusting the variables to their original or optimal state. They are stimulus–response mechanisms in which the response produced reduces the effect of the original stimulus by reversing its direction. For example, if the concentration of a substance in the blood is too high, a negative feedback loop will work to lower the concentration. If the concentration is too low, a negative feedback loop will function to increase the concentration. Most feedback loops in biological systems are negative.

Negative feedback loops are called negative because the information produced by the feedback causes a reversal of the size or effect of the stimulus. Negative feedback loops maintain stability through the action of the nervous or hormonal systems, or both acting together.

A negative feedback loop is shown in Figure 6.1.2 and acts as follows.

- The system is in a stable state.
- A change (stimulus) occurs.
- The change is detected by an appropriate receptor.
- The receptor sends a signal to a control centre (hypothalamus or transmission molecules).
- The control centre sends a signal to an appropriate effector or a specific effector cell, tissue or organ.
- The effector responds to the signal.
- The original state is restored.

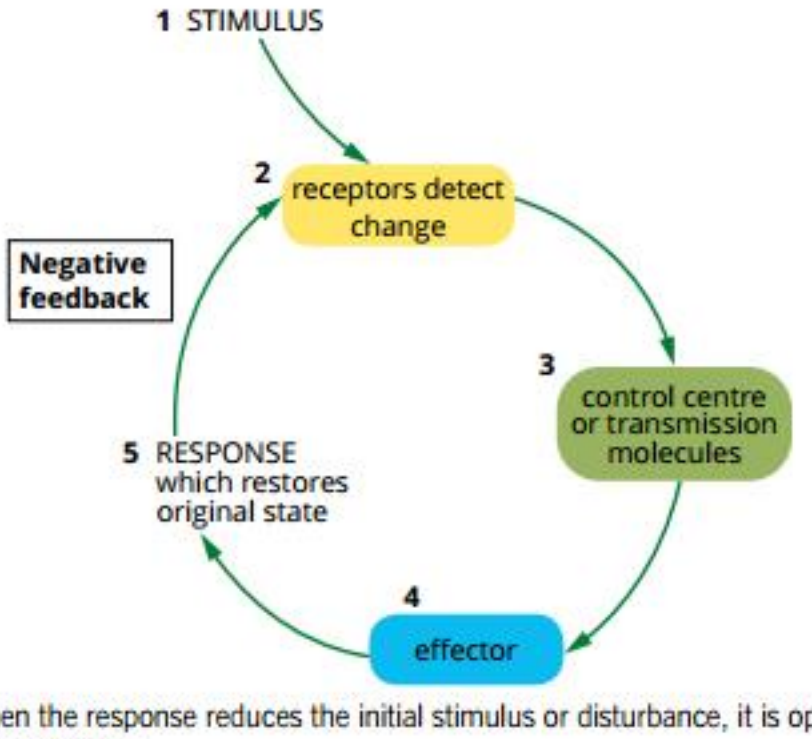


FIGURE 6.1.2 When the response reduces the initial stimulus or disturbance, it is operating as a negative feedback mechanism.

An example of a negative feedback loop is the regulation of blood sugar levels by **insulin** and glucagon. When blood sugar levels increase (stimulus), beta cells (receptors) within the pancreas detect the change and secrete insulin (effector). This lowers the blood sugar levels by stimulating glucose uptake into cells and stimulates the liver into converting excess glucose into the storage molecule, glycogen. Eventually, homeostasis is reached, at which point the pancreas stops releasing insulin. If blood sugar levels then drop too low (stimulus), this is detected by alpha cells (receptors) within the pancreas. Effectors then release glucagon to stimulate glycogen breakdown in the liver into glucose (Figure 6.1.3).

In the control centre, information from sensory receptors is received and compared with a set point (the optimal value for the functioning of that organism). This information is processed with other information about the state of the organism, and an appropriate response is initiated.

Therefore, regulation involves fluctuations around the set point. The size of the fluctuations depends on the:

- sensitivity of the receptor
- tolerance of the control centre to variation from the set point
- efficiency of the effector.

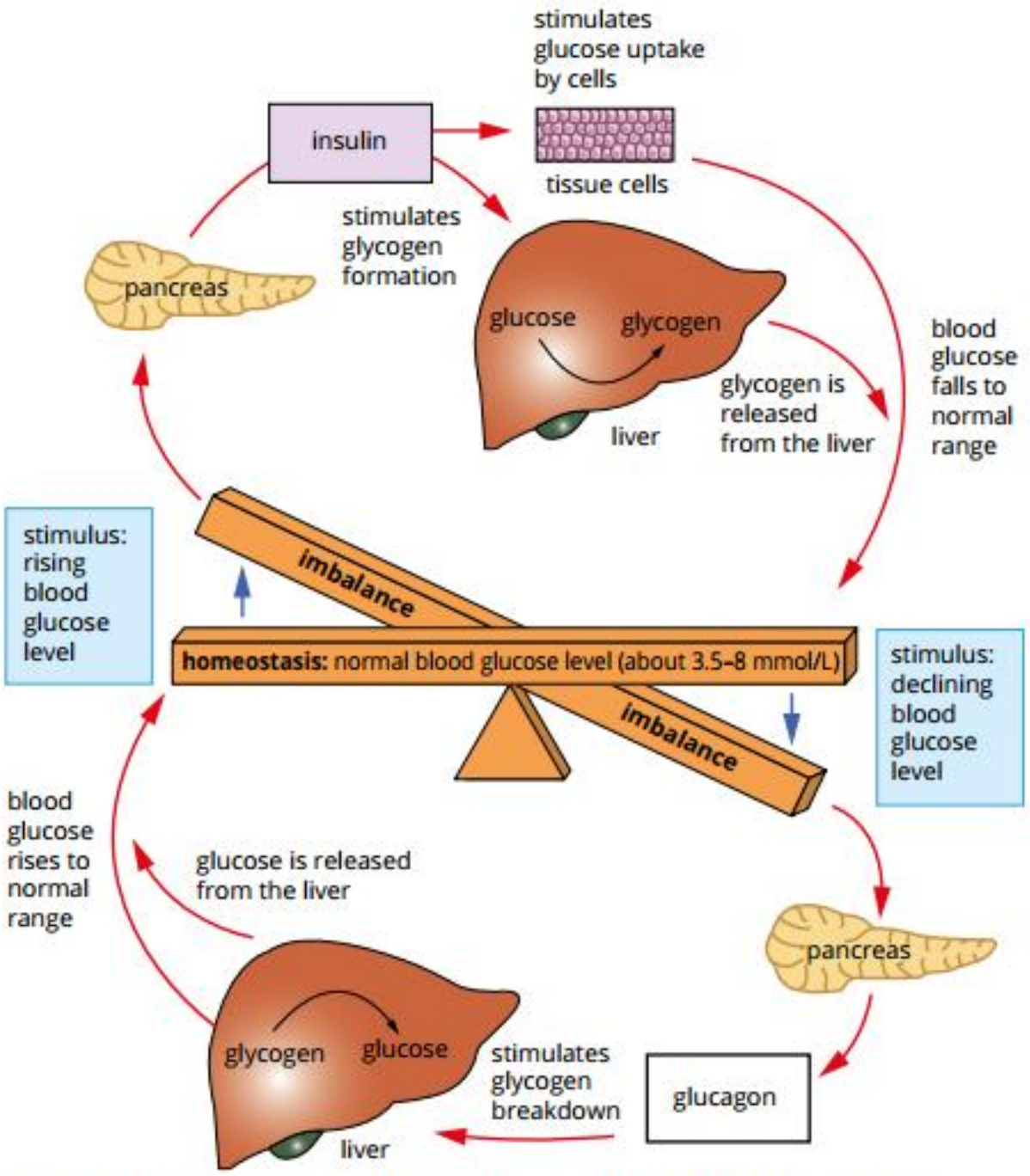


FIGURE 6.1.3 Insulin and glucagon from the pancreas regulate blood sugar (glucose) levels.

Some features of the internal environment, such as blood glucose levels, can vary considerably. Others, such as body temperature in mammals, are tightly controlled.

Effectors can be muscle groups that contract in response to a nerve stimulus, such as the human iris contracting in response to a bright light. Effectors may also be glands that either secrete additional hormones to target muscle groups or release products such as sweat, which has a cooling effect on the body.

In contrast to negative feedback loops, **positive feedback loops** force an organism out of homeostasis by maintaining the direction of the stimulus, and sometimes increasing the stimulus. An example of a positive feedback loop is uterine contractions during childbirth. The hormone **oxytocin** stimulates the uterus to contract.

Rather than the nervous system signalling the endocrine system to lower the oxytocin and reduce the contractions, more oxytocin is produced to stimulate stronger contractions. The contractions work to push the baby into the birth canal and continue until the baby is born and the pressure stimulus is removed.

The differences between positive and negative feedback loops are shown in Figure 6.1.4.

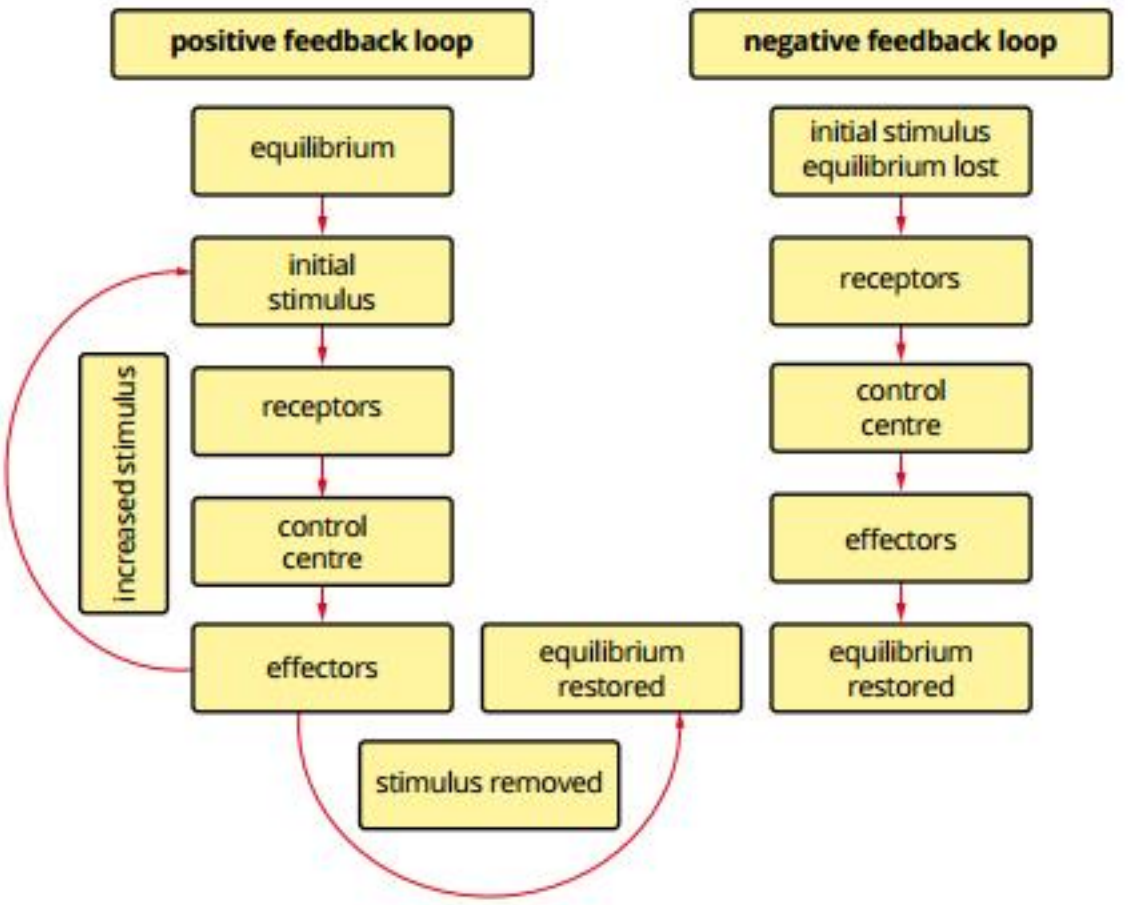
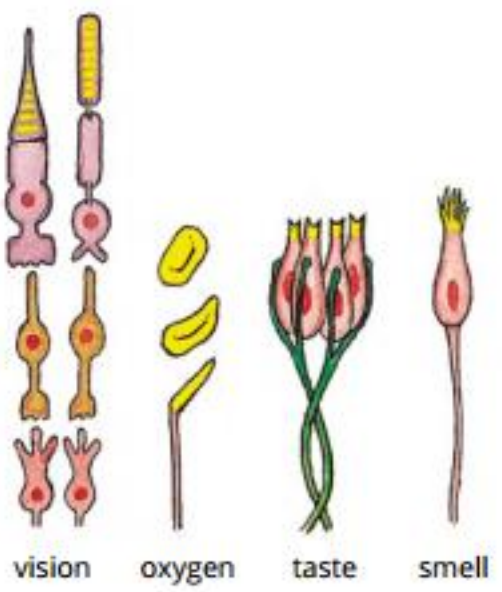


FIGURE 6.1.4 Pathway differences between positive and negative feedback loops



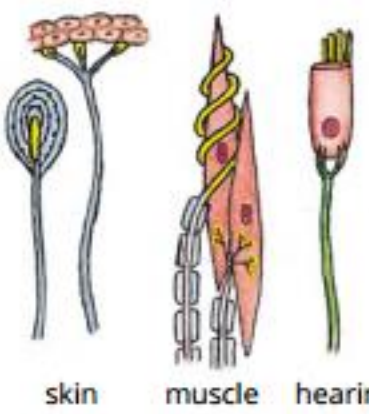


FIGURE 6.1.5 Different sensory receptors. The yellow parts are the regions of the receptor that respond to the particular stimulus. For example; vision (photoreceptor), oxygen (chemoreceptor), taste (chemoreceptor), smell (chemoreceptor), skin (nociceptors), muscle (proprioceptor), hearing (mechanoreceptor)

ANIMAL SENSORY RECEPTORS

All organisms have sensory receptors to detect aspects of either their internal or external environments that may affect their ability to survive and reproduce or to maintain a state of homeostatic equilibrium.

In all animals, including humans, there are receptors which detect changes to internal or external stimuli. Some receptors are located close to the surface of the body and detect stimuli such as pain (nociceptors). Some receptors detect internal states, such as blood pressure and blood chemistry (e.g. oxygen and carbon dioxide levels). From a functional point of view, the types of sensory receptors involved in these senses can be classified as photoreceptors (vision), chemoreceptors (taste, smell, communication), mechanoreceptors (hearing, balance, pressure, touch) and thermoreceptors (temperature). Table 6.1.1 (page 201) and Figure 6.1.5 show some of these receptor types.

Photoreceptors (vision)

Photoreceptor cells that contain light-sensitive pigments appeared very early in animal evolution. Light interacts with the pigment and produces an electrical signal in a sensory nerve. This allows the animal to visually detect changes (stimulus) in its external environment and respond accordingly. The most primitive type of eye consists of a small patch of light-sensitive receptors that acts as a light-detecting organ and cannot form images, for example, the planarian eye-spot shown in Figure 6.1.6.

All vertebrate eyes have an iris, a thin circular disc with a hole, or pupil, in the centre. The iris can contract or relax, and so restricts or enlarges the pupil. Light enters the eye through the pupil and a negative feedback mechanism maintains the intensity of light at an optimal level for the stimulation of the photoreceptors on the retina. If too much light enters the eye, then receptors in the retina will send an impulse to the brain (control centre). A neural impulse will be sent to the iris (effector muscles), causing it to contract and reduce the amount of light entering the eye.

TABLE 6.1.1 Receptor type, examples and stimulus received in complex animals

Receptor type	Examples	Received stimuli
mechanoreceptors	pressure (touch) receptors blood vessels: • baroreceptors	 stretching of blood vessel wall
	skin: • Meissner corpuscle • Pacinian corpuscle • nociceptors	 • light touch • heavy touch • pain
	proprioceptors: • muscle spindles • Golgi cells • joint receptors	 • movement, position of body • gravity • movement with ligaments
	labyrinth in the vertebrate ear: • sacculus and utriculus • semicircular canals • ciliated cells in the cochlear duct	 • gravity and linear acceleration • angular acceleration • sound waves
chemoreceptors	taste buds, olfactory epithelium	specific chemical compounds
thermoreceptors	• thermoreceptors in blood-sucking insects and ticks • nerve endings and receptors in the skin and tongue of many animals	heat
photoreceptors	• eyespots • rods and cones in the retina of the vertebrate eye	light energy

FIGURE 6.1.6 Planarians have simple light-sensitive receptors, called eye-spots, that help them detect and move away from light.

FIGURE 6.1.7 A coloured scanning electron micrograph of the surface of the human tongue. The scale-like projections are filiform papillae, which sense pressure. The red areas are fungiform papillae, which contain the taste buds.

Chemoreceptors (taste and smell)

Chemoreceptors are cells sensitive to different chemicals. They may be grouped inside structures that maximise the probability of detection. Terrestrial animals have specialised organs for olfaction (smell) to detect airborne chemicals, and taste to detect chemicals in food and drink (such as the taste buds on the tongue shown in Figure 6.1.7).

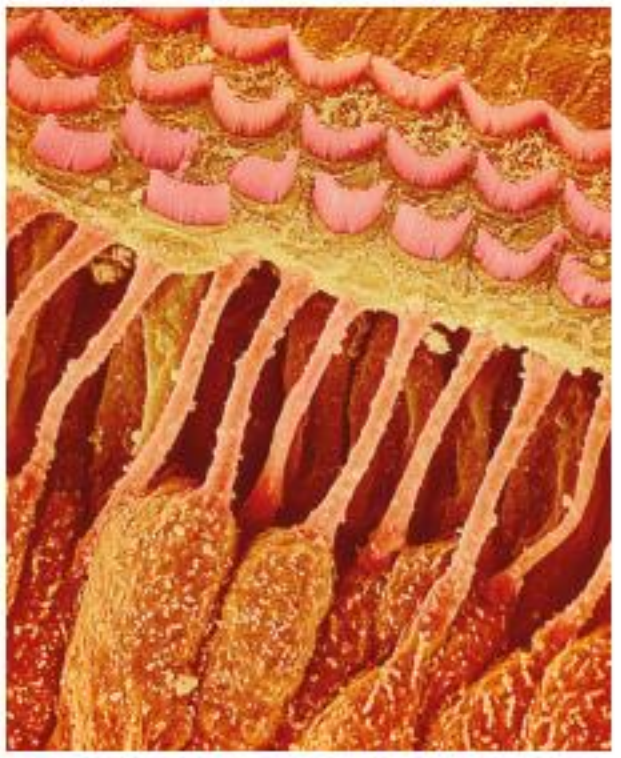


FIGURE 6.1.8 A coloured scanning electron micrograph of sensory hair cells in the cochlea. The inner ear converts sound waves into nerve impulses.

Mechanoreceptors (hearing, touch and pain)

Sound travels as vibrations through air, water and solids. Animals detect sound using mechanoreceptors, which are **sensory neurons** that can detect minute vibrations. Vibration-sensitive neurons may be attached to larger vibrating structures that select and filter, and sometimes amplify the frequencies that are important to the animal. In human ears, vibrations are amplified and are then transmitted to a fluid-filled canal (cochlea) with sensory hair cells. You can see sensory hair cells in Figure 6.1.8. Vibrations entering the inner ear generate nerve impulses that travel to the brain along the auditory nerve. The inner ear can transmit information about the volume and pitch of a sound. If sound wave pressure falls within an acceptable range, a state of equilibrium will be in place. However, if the sound pressure is too great, then a negative feedback response will be initiated where the animal will respond behaviourally by blocking out the sound stimulus, or by moving away from it.

Mechanoreceptors detect external stimuli, including pressure and touch. Figure 6.1.9 shows some of these receptors in skin. A range of receptors detect internal mechanical stimuli, including joint position, muscle tension, and tension in the walls of organs such as the lungs and stomach. Examples of mechanoreceptors are stretch receptors, which aid in proprioception (the unconscious perception of body positioning and movement). When you stand on one foot, the slight movements you feel in your ankle are your body correcting its position because of the stimuli detected by these stretch receptors.

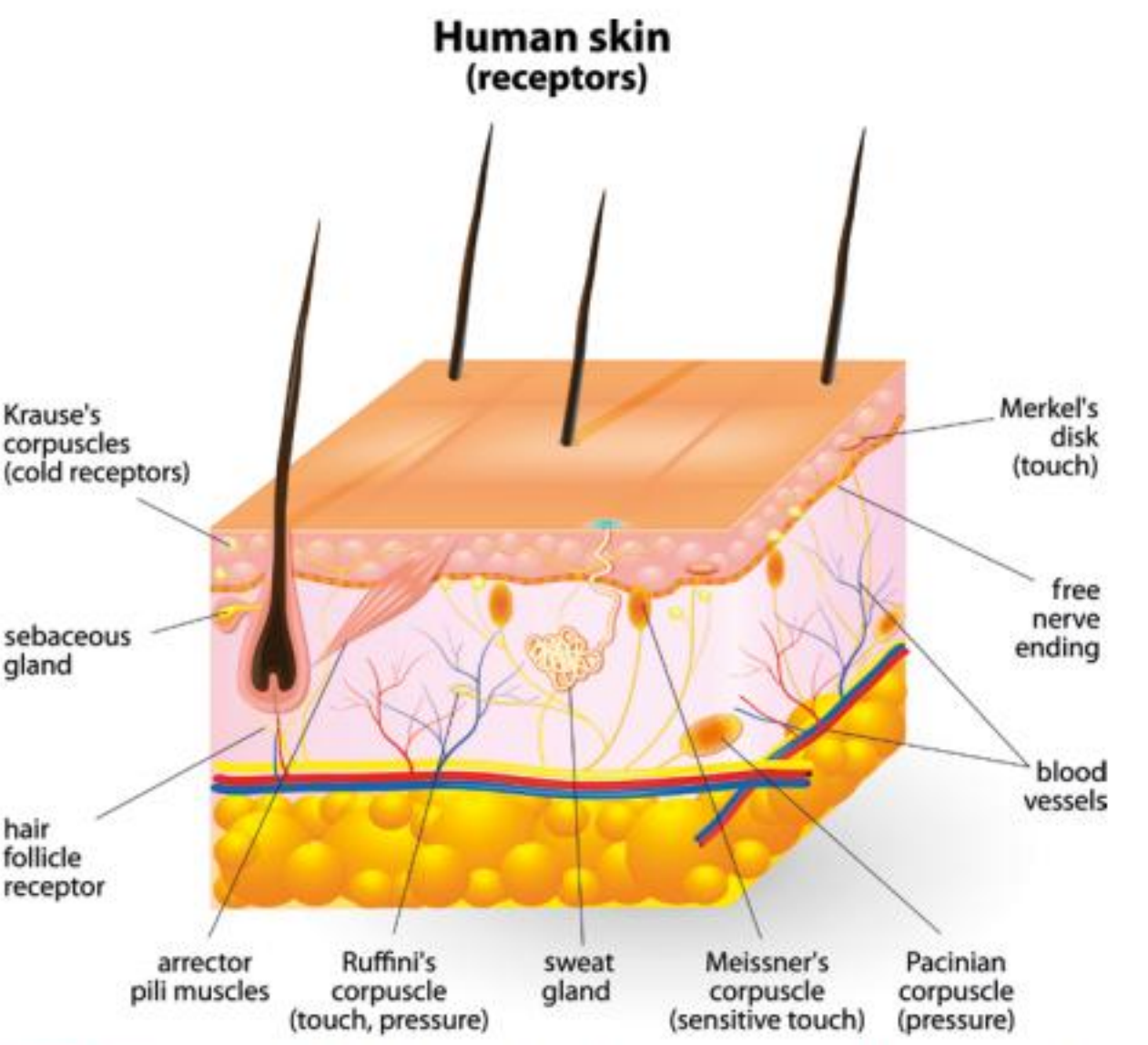


FIGURE 6.1.9 A cross-section of human skin showing the various receptors for touch, pressure and temperature.

The process of pain transmission is called nociception, and pain receptors (nociceptors) are found in most body tissues. The homeostatic response to a tissue injury, such as a cut on the finger, is very complex and involves the release of a range of chemicals that will initiate inflammation of the local tissue, increase blood flow, start the blood-clotting response and cause white blood cells to move to the injury site. It is a series of both positive and negative feedback loops that are activated and deactivated at various times during the initial response and healing process.

Thermoreceptors (temperature)

Thermoreceptors detect changes in heat energy. Thermoreception is complex and involves the detection of heat energy as well as changes in temperature. Different parts of the body have more receptors than others. For example, your hand, has more thermoreceptors than your leg, which means that it is more sensitive to temperature changes.

6.1 Review

SUMMARY

- Homeostasis is the maintenance of variables in a system within certain limits.
- Animals have a range of internal and external sensory receptors. These include mechanoreceptors (touch, hearing and nociceptors for pain detection), chemoreceptors (taste and smell), thermoreceptors (heat), and photoreceptors (vision).
- Endocrine (hormone) and nervous systems have important roles in negative feedback mechanisms that promote homeostasis in animals.
- Negative feedback loops are stimulus–response mechanisms that respond to changes in the body by adjusting variables back to their original or optimal state, thus reversing the direction of the stimulus.
- Most feedback loops are negative and contain a stimulus, receptor, control centre, effector and response component.
- Positive feedback loops are the opposite of negative feedback loops. They promote a process rather than reversing the effect of the stimulus.

KEY QUESTIONS

Describe

1 Complete the table below by recalling and placing each of the following stimuli next to the receptor that it responds to: pain stimulus, temperature, touch and pressure, chemical stimulus, blood pressure.

Receptor	Stimulus
baroreceptor	
chemoreceptor	
mechanoreceptor	
nociceptor	
thermoreceptor	

2 Recall each part of the stimulus-response model. Arrange the following terms from first (A) to last (E) in the order of their involvement in a physiological response: receptor, response, control centre, effector, stimulus

A	
B	
C	
D	
E	

continued over page

6.1 Review *continued*

Apply

3 Define 'homeostasis' and explain why it is important.

4 What is the difference between nervous system response and endocrine system response in homeostasis?

5 Compare the processes of negative and positive feedback.

6 Explain, using an example, how negative feedback loop functions.

7 Identify the role of the control centre in the homeostatic stimulus–response model.

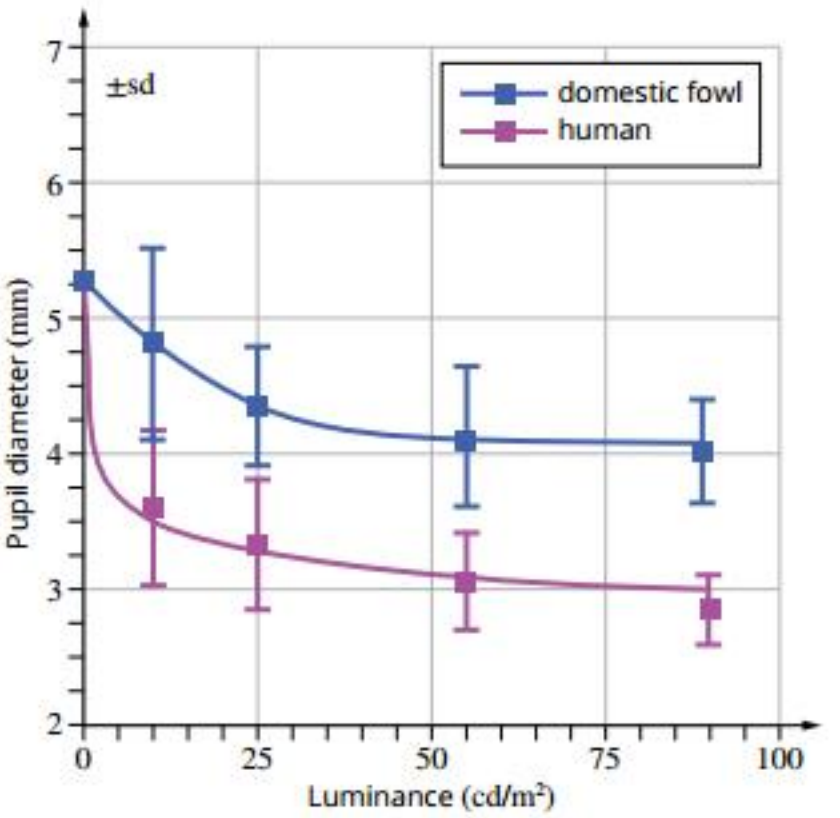
Analyse

8 Imagine you are walking in your garden picking flowers, when a thorn on a rose bush pricks your arm. You immediately pull your arm back.

- **a** Identify the receptor in this stimulus–response model and describe what this receptor is detecting.
- **b** Identify the effector in this stimulus–response model and describe what this effector does.
- **c** Determine if the effector outlined in this stimulus–response model is a muscle or a gland.

9 Different animals have the ability to dilate or constrict the iris of their eye to regulate the amount of light entering. The graph below shows the pupil diameter (nm) of a human and a domestic fowl at different levels of light (luminance).

- **a** Analyse the data within the graph and describe the relationship seen in the effector muscles of the human pupil when the level of light is increased.
- **b** Provide a reason as to why the domestic fowl has a larger pupil diameter across all light levels compared with the human.



6.2 Neural control pathways

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- describe the structure and function of nerve cells, including dendrites, soma, body, axon, myelin sheath, nodes of Ranvier, axon terminal and synapse
- distinguish between sensory neurons, interneurons and motor neurons
- explain the passage of a nerve impulse in terms of transmission of an action potential and synaptic transmission, referring to neurotransmitters, receptors, synaptic cleft, vesicles, postsynaptic and presynaptic neurons and signal transduction.

Organisms receive information about their environment and respond to external stimuli through the network of neural pathways that makes up the nervous system. The nervous system works closely with the endocrine system to respond to environmental changes and regulate the internal environment.

The rapid responses characteristic of most organisms are brought about by the nervous system. Nervous systems typically involve a more direct pathway of communication between parts of the body than hormonal responses.

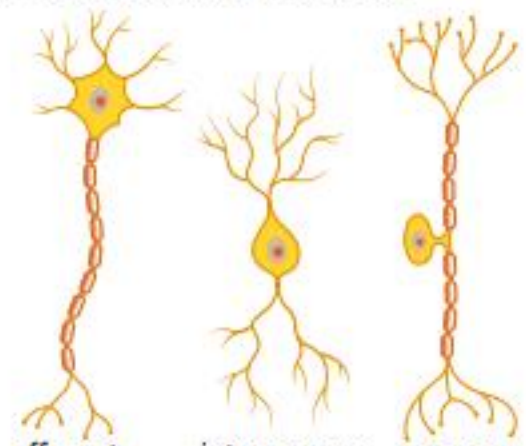
NEURONS

The neuron (or nerve cell) is the functional unit of nervous systems. Neurons are specialised cells with structures that enable rapid transmission of information between cells. As shown in Figure 6.2.1, there are three basic functional and structural classes of neurons.

- **Afferent (sensory) neurons** communicate information from tissues and organs to the **central nervous system (CNS)**. Sensory neurons have **dendrites** on both ends. One of these dendrite bundles is associated with specialised sensory receptors. Sensory neurons also have a **cell body** (or **soma**) centrally located along the **axon**, which contains organelles that keep the neuron functioning.
- **Interneurons** conduct nerve impulses within the nervous system and transfer signals between sensory and motor neurons. Interneurons can also communicate with each other, forming circuits of various complexity. Interneurons have short dendrites and a long or short axon. Most, but not all, interneurons lack a **myelin sheath**.
- **Efferent (motor) neurons** transmit information from the CNS to the tissues and organs (effector cells). Motor neurons have dendrites and a cell body or soma located in the CNS and an axon located outside of the CNS.
- The basic structure and function of neurons is very similar across all groups of the animal kingdom. Figure 6.2.2 on page 206 shows the basic neuron structure.

These different types of neurons have different structures but are made up of the same basic components: one or more dendrites, a cell body (soma) and an axon (Figure 6.2.2, page 206). Branching dendrites receive signals from other cells and transmit these to the cell body. A single axon conducts a signal from the nerve cell body to nerve endings, which form **synapses** with other cells.

a Functional classes of neurons



b Structural classes of neurons

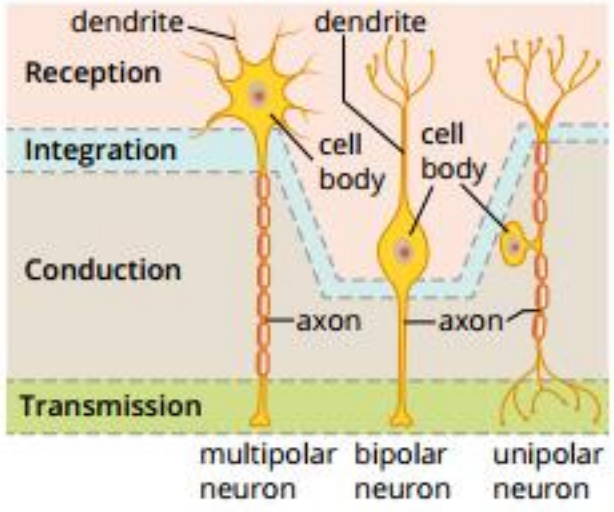


FIGURE 6.2.1 Different types of neurons vary in structure according to their function and can be divided into classes based on their (a) function or (b) structure.

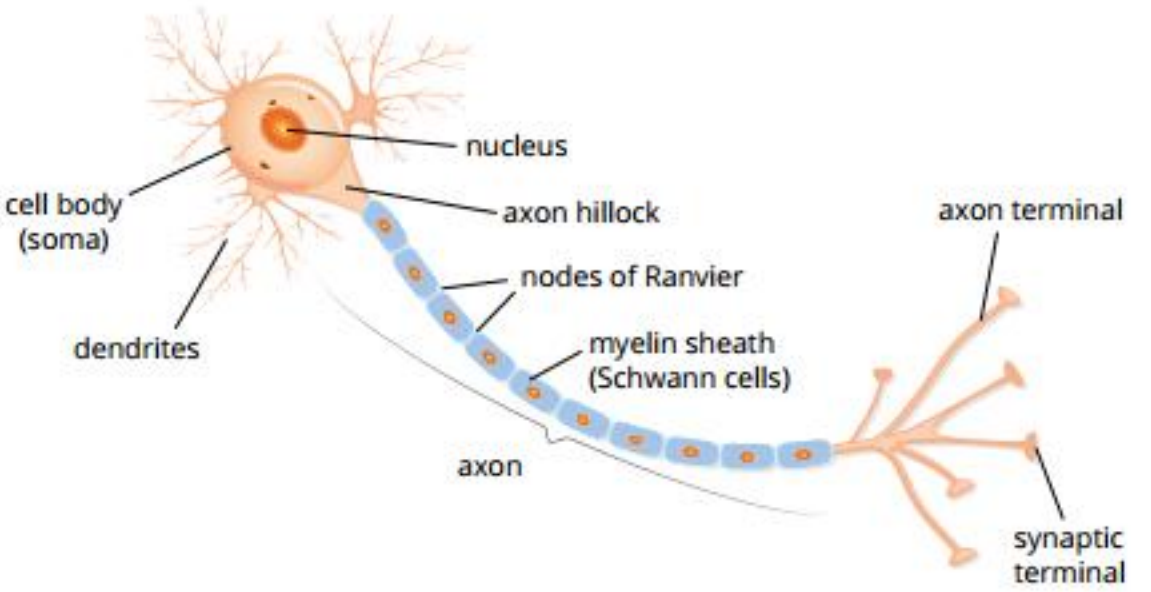


FIGURE 6.2.2 The key structures of a generalised neuron are very similar across all groups of the animal kingdom.

Longer axons may be covered with a myelin sheath. Myelin is a protein- and lipid-rich substance produced in the CNS by Schwann cells. A series of Schwann cells wraps tightly around the axon. The gaps between the Schwann cells are called the **nodes of Ranvier** (Figure 6.2.2). While the myelin sheath acts as an electrical insulator for the axon, the nodes are rich in ion channels (Figure 6.2.3). This allows the nerve signal (**action potential**) to 'jump' from node to node, instead of having to move more slowly along the whole of the axon membrane. Generally, longer sensory neurons will be myelinated, and other neurons, which have no need for fast reflex action (such as interneurons), will be non-myelinated.

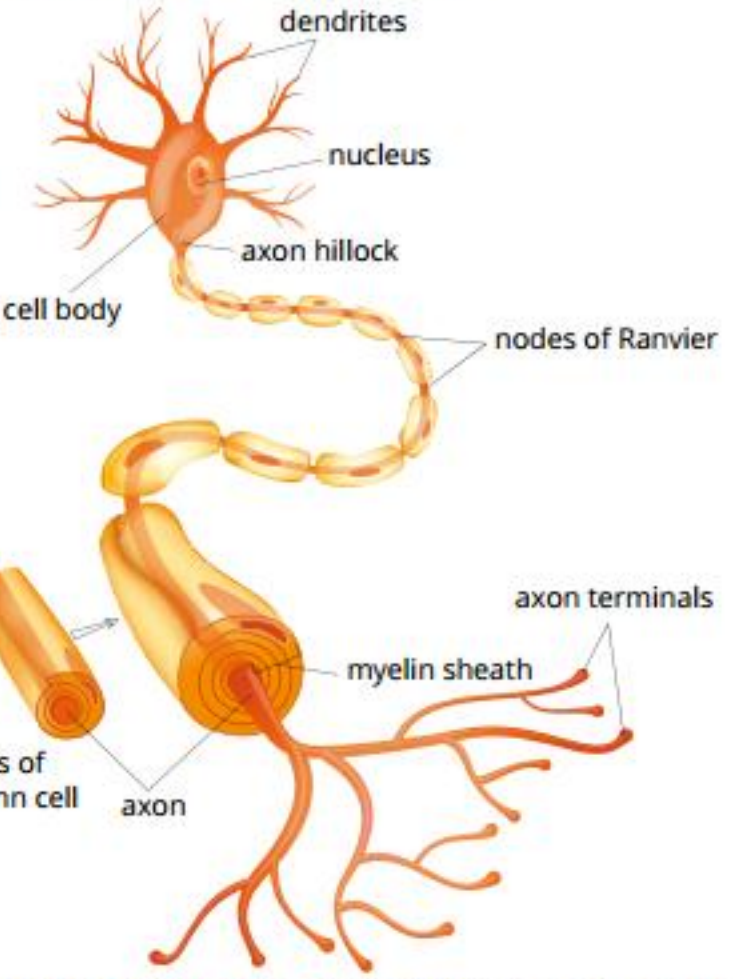


FIGURE 6.2.3 Myelin sheath surrounding a neuron is produced by Schwann cells. The axon of some neurons is sheathed within a lipid- and protein-rich substance called myelin.

Neurons are also grouped into classes based on their structure: multipolar neurons have two or more dendrites that are separate from the axon; bipolar neurons have a single dendrite; and unipolar neurons have just one extension from the cell body, made up of an axon and dendrite.

In nervous systems, nerve cells connect to form pathways between receptor cells and responsive organs, sometimes via a central organ such as the brain or spinal cord. For example, when sensory cells detect an environmental disturbance, a signal is generated that passes as an electrical message along two or more neurons to reach particular effector cells, such as muscle or gland. Chemical transmitters communicate from sensory cell to nerve cell, between nerve cells, and from nerve cell to the effector cell that produces the response. You can see this in Figures 6.2.8 and 6.2.9 on page 211.

Although at times the passage of an impulse along a neuron has been likened to an electrical current flowing in a wire, there are great differences between the two processes. The nerve impulse travels along the surface of the membrane (which has a high impedance to charge) and uses chemical messengers to bridge the gap between neurons. The purely electrical charge travels at a much faster speed within the continuous section of wire, with any gaps effectively stopping the passage of the electrical current.

Neural response pathways are highly specific. Receptor cells are sensitive to specific environmental disturbances: the 'wiring' of neuronal pathways is direct, and to respond, effector cells must possess specific receptors. Because of the directness of the pathway, and the speed of conduction along nerve fibres, control by nerves is usually extremely rapid and short in duration, and the response is precisely directed.

PHYSIOLOGY OF A NERVE IMPULSE

Nervous responses require more energy than hormonal responses do. In nervous systems, the passage of signals (**signal transduction**) along nerve axons requires considerable energy to re-establish the balance of ions across the axon membrane after each signal, while hormones are carried to their target cells in the circulatory system (see Module 6.3 217).

Neurons establish steep concentration gradients of ions across their cell membranes to enable an electrical impulse or action potential to occur. Establishing these gradients uses active transport, which requires a large amount of energy. Therefore, neurons need many mitochondria and a large supply of glucose.

Signal transduction in neurons

Transduction of a signal carried by the nervous system uses a combination of electrical and chemical signalling involving the opening of gated ion channels and the release of **neurotransmitters** through exocytosis.

Signal transduced into a neuron

When a neuron is stimulated, gated sodium and potassium ion channels on its membrane are opened. The sudden movement of the ions into and out of the neuron initiates the action potential, which travels the length of the axon of the neuron in a wave-like sequence, opening other ion channels along the membrane. An action potential is the reversal of the normal potential difference across a cell membrane, or between the inside and the outside of a nerve fibre.

Action potential

An action potential or nerve impulse is a wave of electrical change that passes rapidly along an axon membrane. An action potential begins when a stimulus disturbs the cell membrane on a dendrite, causing sodium ion channels to open. When a neuron is not stimulated or is at rest, the membrane is **polarised**—the inside of the cell is more negative.

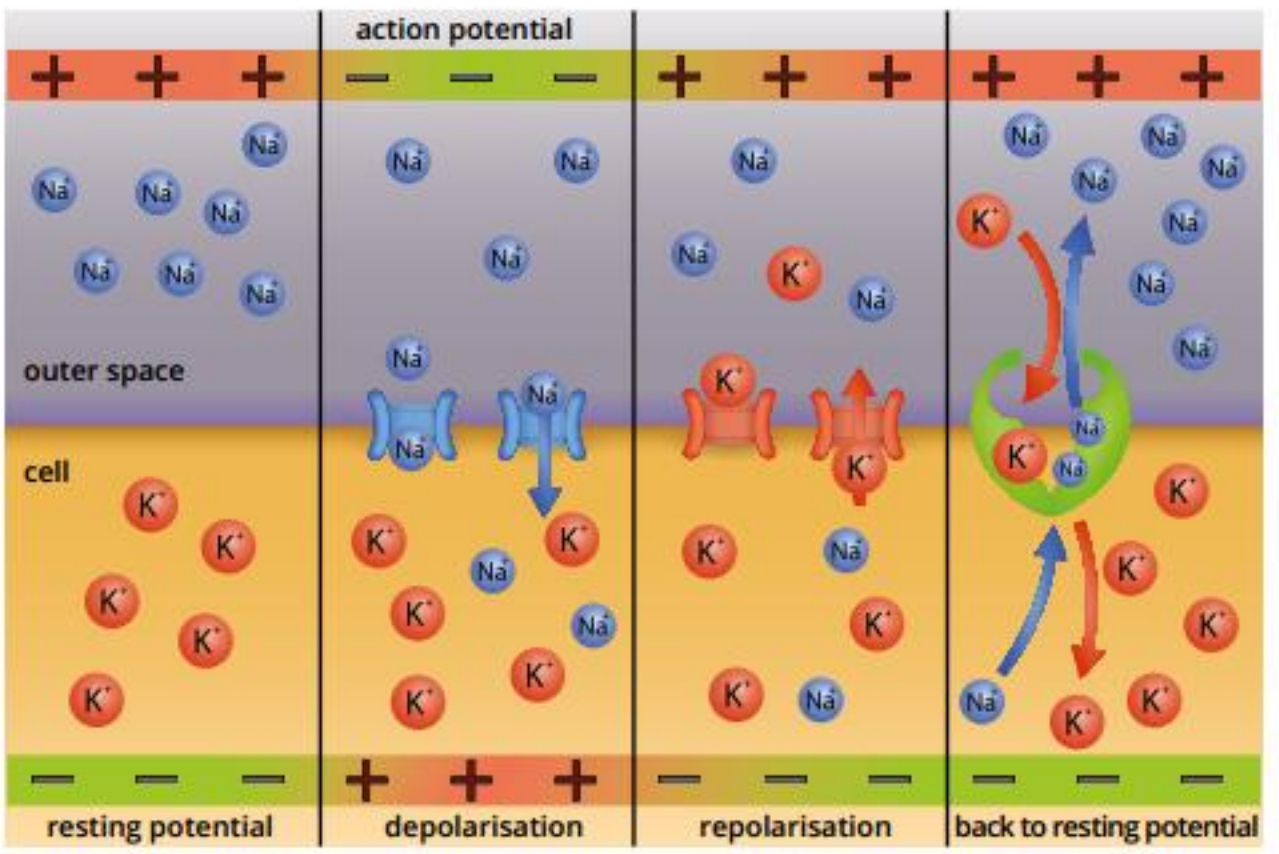


FIGURE 6.2.4 The inside of the neuron is negative with respect to the outside of the neuron, and there is an unequal distribution of sodium ions (Na^+) and potassium ions (K^+) across the cell membrane. To the left depolarisation occurs as the action potential moves along the membrane and Na^+ ions flood into the cell via Na^+ ion channels. Repolarising takes place as K^+ ions flood out of the cell through K^+ ion channels. The resting potential is again restored only by using ATP to actively pump Na^+ ions back out of the cell and K^+ ions into the cell, as seen at the right of the image. This requires large amounts of energy and so neurons will possess large numbers of mitochondria.

A charge difference is maintained between the inside and outside of the cell largely by active transport using sodium–potassium pumps. The pumps actively transport sodium ions out of the cell and potassium ions into the cell (Figure 6.2.4).

When an action potential is initiated, and sodium ion channels are stimulated to open sodium ions (Na^+) flow into the neuron. Because sodium ions are positively charged, their influx causes the inside of the neuron to become slightly less negative (more positive). This causes the membrane to become depolarised. If the depolarisation is sufficient to reach the threshold potential for the cell, Na^+ channels in the membrane open and Na^+ floods into the cell along its concentration gradient, initiating an action potential (Figure 6.2.5). Because sodium ions are positive, the inside of the cell becomes briefly positive, which causes potassium ion (K^+) channels to open, and K^+ diffuses out of the cell along its concentration gradient. This process takes approximately 4 ms.

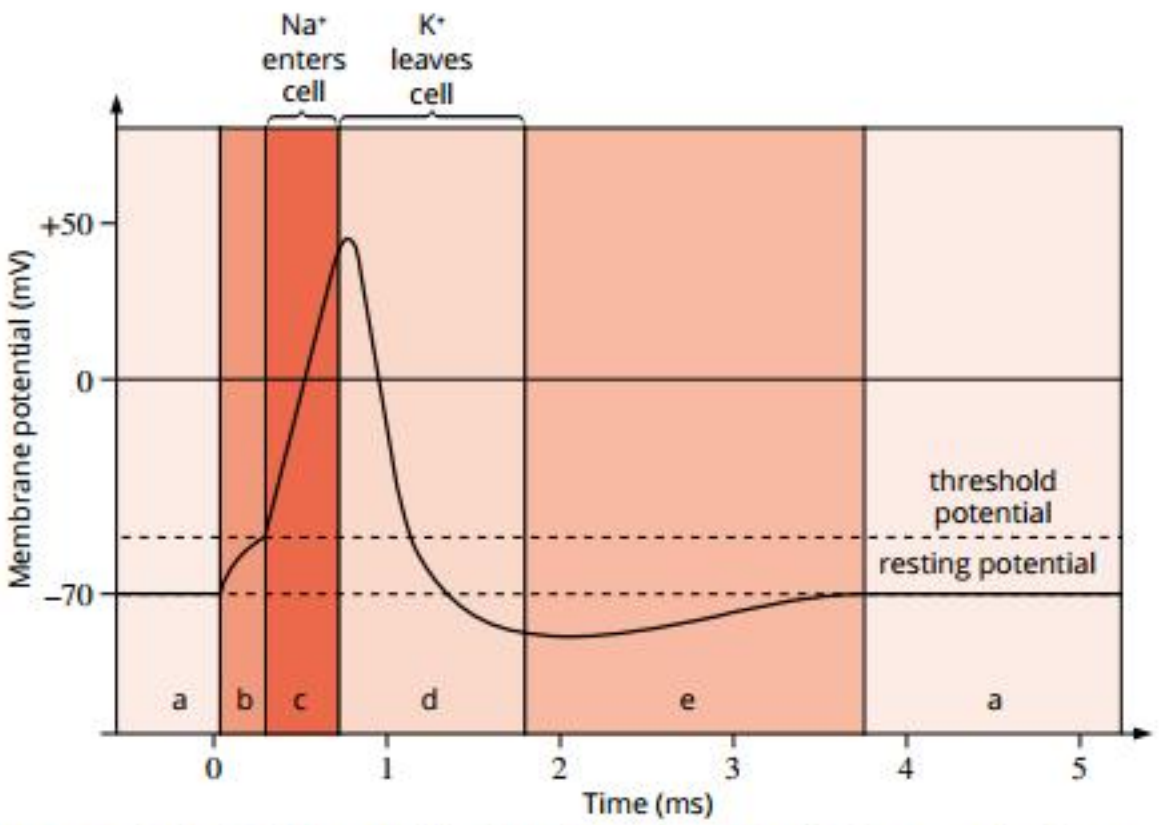


FIGURE 6.2.5 A simplified diagram of the internal membrane potential of a nerve cell during an action potential: (a) resting membrane potential' (b) depolarisation reaches the threshold potential of the axon, (c) the membrane depolarises and the inside of the cell becomes briefly positive as sodium ions (Na^+) diffuse into the cell, (d) the potential becomes negative again as potassium ions (K^+) diffuse out of the cell, (e) the resting membrane potential and the original distribution of ions are re-established by active transport of Na^+ and K^+ across the membrane.

As the positive potassium ions leave, the inside of the cell becomes negative again. Therefore, the inside of the cell first becomes briefly positive and then negative again. These electrical changes are the action potential. Following the action potential, the Na^+ and K^+ channels close and the membrane returns intracellular ion concentrations to their initial values. However, for the resting potential to be reinstated, Na^+ ions must be moved out of the cell and K^+ ions moved in—both against their concentration gradients. To achieve this, the membrane is studded with embedded Na^+–K^+ pumps, which use ATP energy to physically pump the ions against the concentration gradient. This is why neurons contain more mitochondria than other somatic cells in the body.

Once an action potential is generated, it sweeps along the neuron by conduction. During an action potential, when the sodium ions diffuse into the cell, some diffuse sideways inside the axon, depolarising the adjacent region of the axon membrane. As with the initial stimulus, this causes Na^+ channels in this part of the membrane to open and Na^+ floods into the cell, initiating an action potential in this region, as shown in Figure 6.2.6. This depolarises the next adjacent region of the axon membrane, and so the cycle continues, conducting the action potential along the length of the axon membrane.

As the action potential moves along the membrane, there can be some leakage of Na^+ ions from the axon, slowing down the rate at which the action potential passes along the axon membrane. The presence of a myelin sheath along the axon prevents such leakage and greatly increases the speed of the action potential along the axon. Na^+ channels reside mainly in the regular gaps (nodes of Ranvier) in the myelin sheaths.

ATP (adenosine triphosphate) is a molecule that carries the energy required for cellular processes.

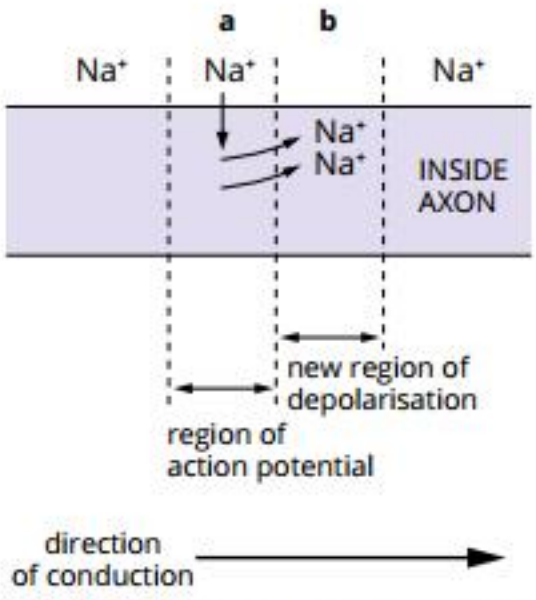


FIGURE 6.2.6 (a) As Na^+ enters the cell, some diffuses sideways, (b) depolarising the adjacent region of membrane and causing an action potential in this region. In this way, an action potential is conducted along the axon.

Cellular response of a neuron

Vesicles containing the neurotransmitters are located at the **synaptic terminals**. When the action potential eventually reaches the end of the axon, the synaptic terminals, it causes another ion, calcium (Ca^{2+}), to enter the cell. The increased concentration of Ca^{2+} causes these vesicles to fuse with the nearby membrane (known as the presynaptic membrane) and to release the neurotransmitters into the synaptic cleft via exocytosis.

The synaptic cleft is the space between the presynaptic cell and the postsynaptic cell. It is sometimes referred to as the synaptic gap.

The postsynaptic cell can be another neuron or another type of target cell such as a muscle or gland cell. The neurotransmitters released from the synaptic terminal diffuse across the synaptic cleft and bind to specific receptors on the postsynaptic cell. If the postsynaptic cell is another neuron, binding of neurotransmitters to the receptors on its dendrites triggers the ion channels of the postsynaptic membrane to open, leading to release of neurotransmitters from the synaptic terminals (Figure 6.2.7). In this case, the signal has been transformed, or transduced, from an electrical impulse into chemical signalling molecules and back again. If the postsynaptic cell is another type of cell, then it will respond to the action potential as an effector cell or tissue. For example, a bundle of muscle fibres will contract, or endocrine tissue will secrete hormones. Excess neurotransmitters are either broken down by enzymes or drawn back into the presynaptic cell by receptor channels or carrier molecules.

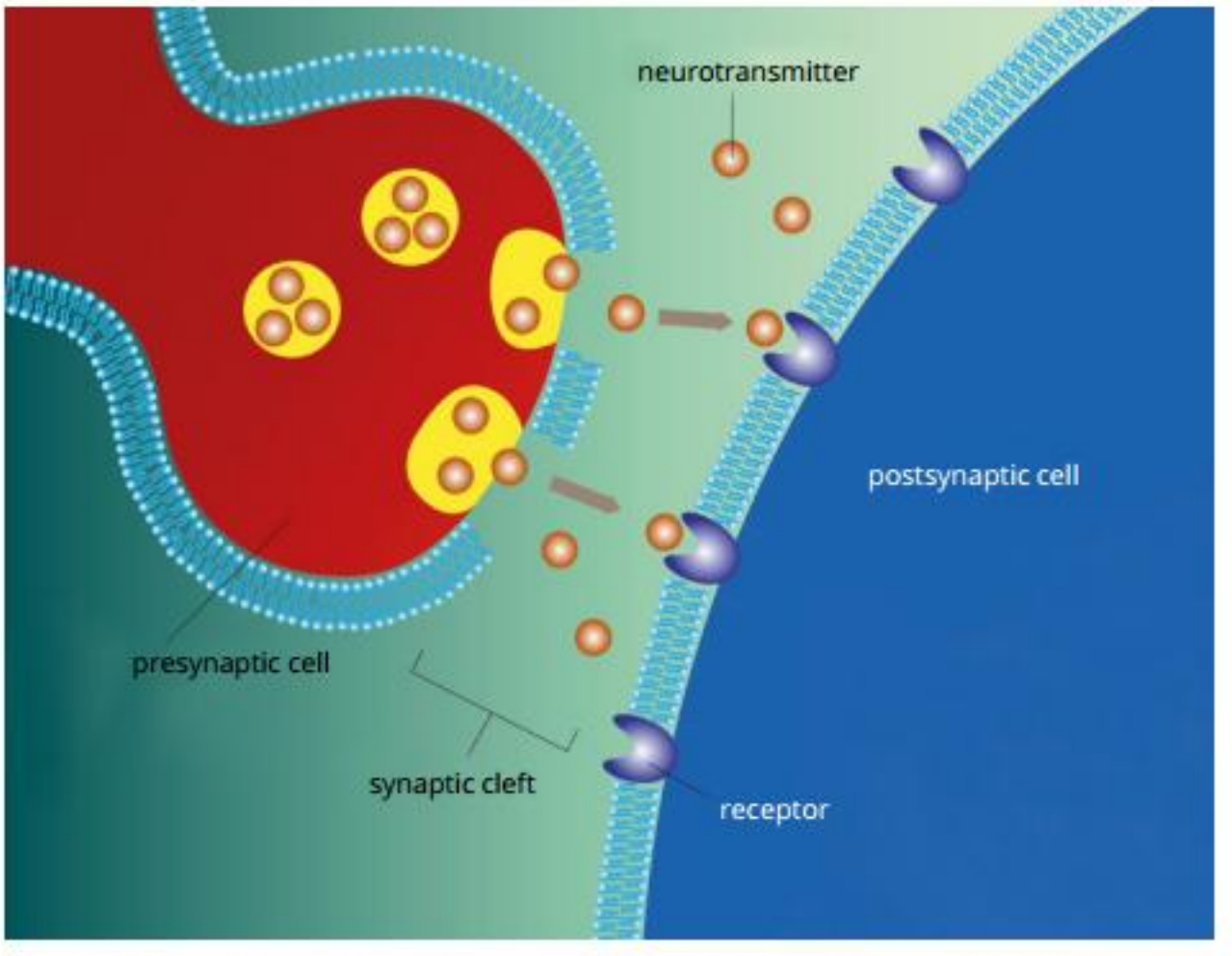


FIGURE 6.2.7 Secretory vesicles containing neurotransmitters fuse onto the presynaptic membrane, releasing the neurotransmitters into the synaptic cleft. Neurotransmitters diffuse across the cleft and bind to receptors found on the postsynaptic cell

Reflex responses

The ability to detect and quickly respond to changes in the internal and external environments is fundamental to the survival of all animals. Some responses to stimuli happen very quickly. If you put your hand on a hot stove or tread on a pin, your body will react in a rapid and coordinated way to move away from the painful stimulus. Stimulation of various receptors in the skin (thermoreceptors for temperature and nociceptors for pain) sends a message along nerves to the spinal cord, where the message is passed on to nerves that stimulate the muscles involved in withdrawing your hand (Figure 6.2.8) or pulling away your foot. At the same time, other nerves in the spinal cord carry the message up to your brain and you become aware of the pain. But, before you are aware of the pain, you have pulled your foot or hand away. This is an example of a rapid, unconscious response called a **reflex**. This kind of reflex protects the body from further injury. The contraction and dilation of the iris in response to light is also a reflex response.

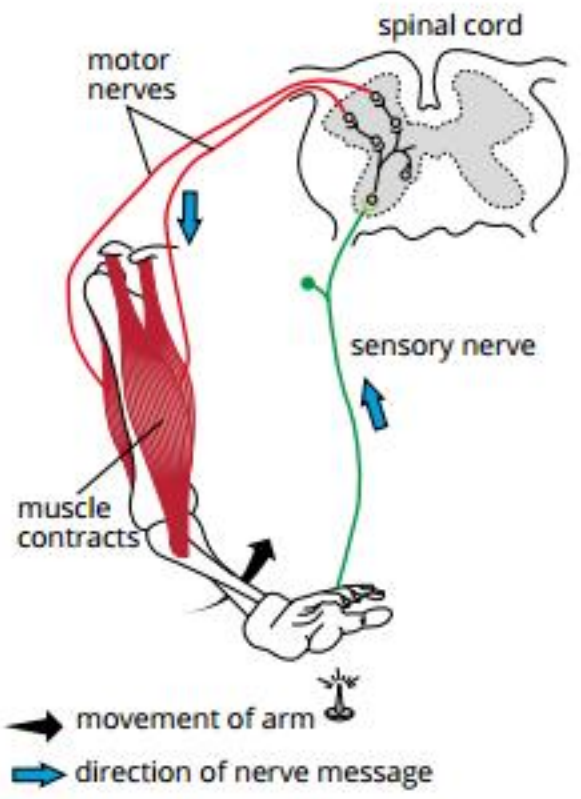


FIGURE 6.2.8 The pathway involved in the reflex withdrawal of the hand in response to a painful stimulus

A reflex is the simplest type of nervous response in an animal, and it may involve just two or three cells. A sensory receptor detects a change and when it has received enough stimulation, it sends a signal via a sensory (or afferent) neuron to the spinal column (part of the CNS). Here it will interact with a motor (effector) neuron to send an impulse to cause muscle fibres to contract—the reflex response. The response is unconscious and automatic; it is not modified because of information received from other parts of the body. The nerve net of a sea jelly produces only reflex responses. Reflex responses are also important in highly complex animals, such as vertebrates, because they occur rapidly and unconsciously—for example, defensive reactions and changes in posture.

The knee-jerk (stretch) reflex is an example of the simplest form of reflex in mammals and involves only two neurons. It is known as a monosynaptic reflex. Most reflexes are polysynaptic and involve one or more additional neurons called interneurons (Figure 6.2.9) in the CNS, between the sensory and motor nerves, as in the withdrawal reflex.

Reflexes are also involved in homeostatic regulation of systems such as the circulatory system. For example, the baroreceptor–heart rate reflex helps maintain blood pressure in mammals. An increase in blood pressure increases the stretch on baroreceptors in major arteries, causing an increase in impulse activity from these sensory neurons. This leads to a reflex decrease in heart rate and a consequent decrease in blood pressure.

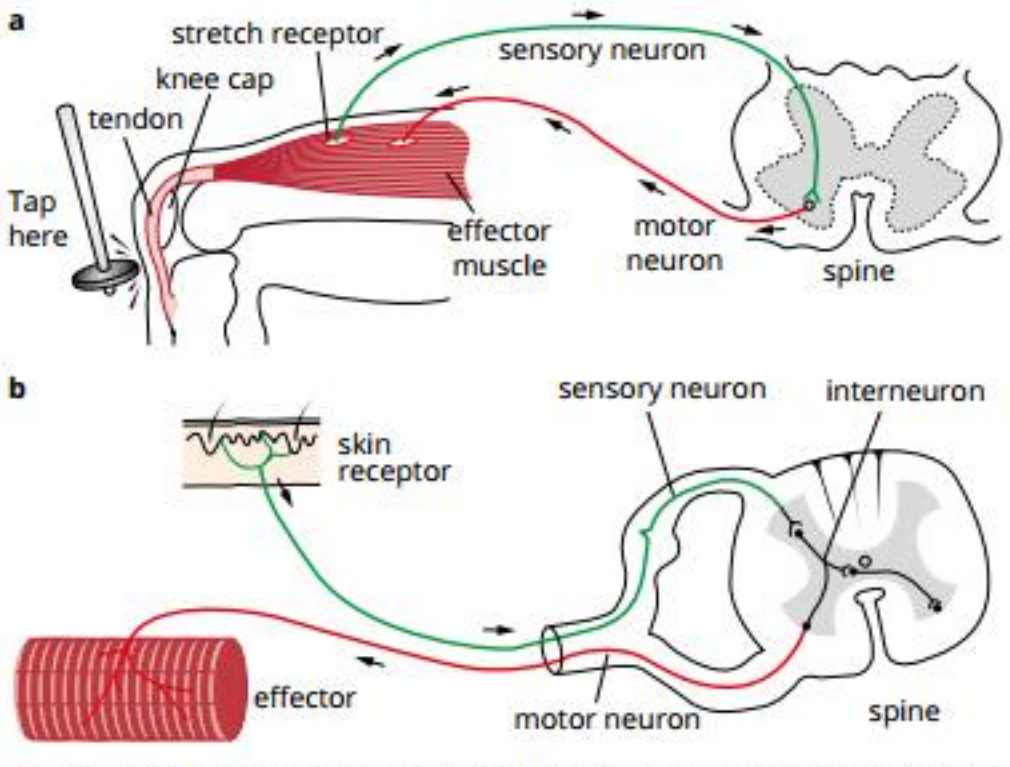


FIGURE 6.2.9 (a) The knee-jerk response is an example of a monosynaptic reflex. It is used most frequently by doctors to detect potential damage to motor areas of the CNS. (b) The withdrawal reflex is triggered by pain receptors in the skin and involves interneurons.

If you concentrate, you can increase or decrease your knee-jerk response. You can also repress the reflex that would cause you to remove your hand from a painful or uncomfortable stimulus. This indicates that there is a connection from the brain that can consciously override the automatic response. You can consciously send signals down the spinal cord to prevent the reflex stimulation of muscles by the sensory nerves. For example, most people do not pull away when being given an injection.

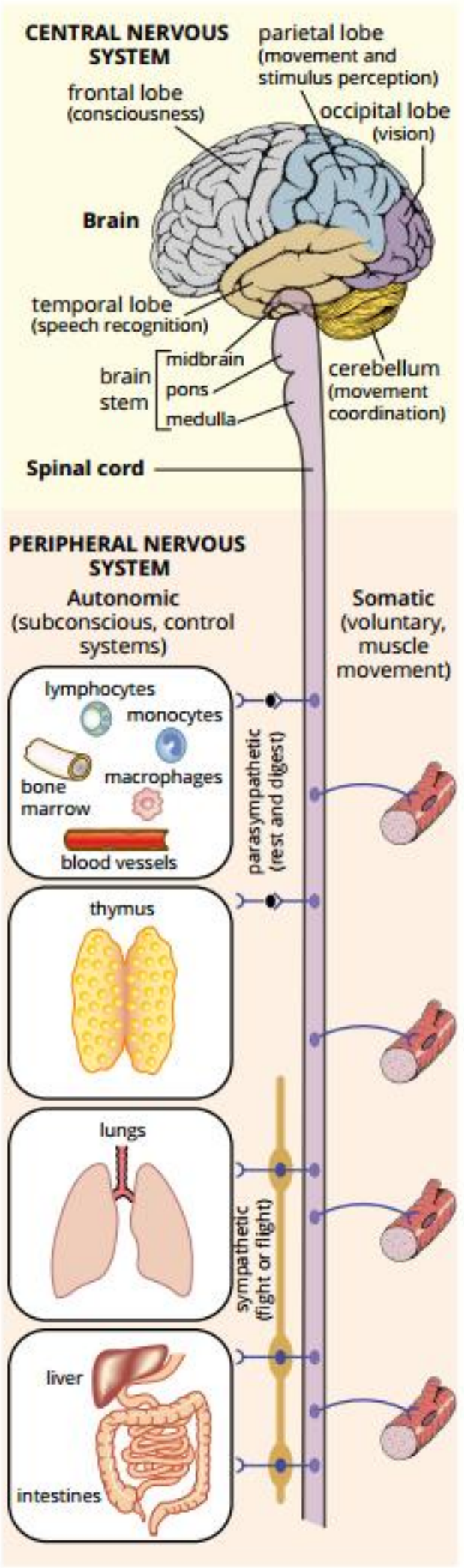


FIGURE 6.2.11 The nervous system of vertebrates consists of a brain and a spinal cord. The CNS is made up of the brain and spinal cord. The peripheral nervous system is a network of nerves that branch throughout the body and send signals to and from the CNS and comprises the autonomic and somatic nervous systems.

Interneurons and integration

An interneuron is a neuron that transmits information from one neuron to another. When interneurons are part of a pathway, the opportunity for coordination and integration increases. For example, the movements of joints operate by the action of opposing sets of muscles: one set causes flexion (bending), the other causes extension (straightening). Your elbow is flexed by the action of the biceps muscle, and extended by the triceps muscle, as shown in Figure 6.2.10. When a muscle is stimulated to contract, such as in a reflex response, interneurons in the spinal cord also carry a message inhibiting the contraction of the opposing muscle. For example, this prevents both the biceps and triceps from contracting at the same time, which would put the elbow under severe strain.

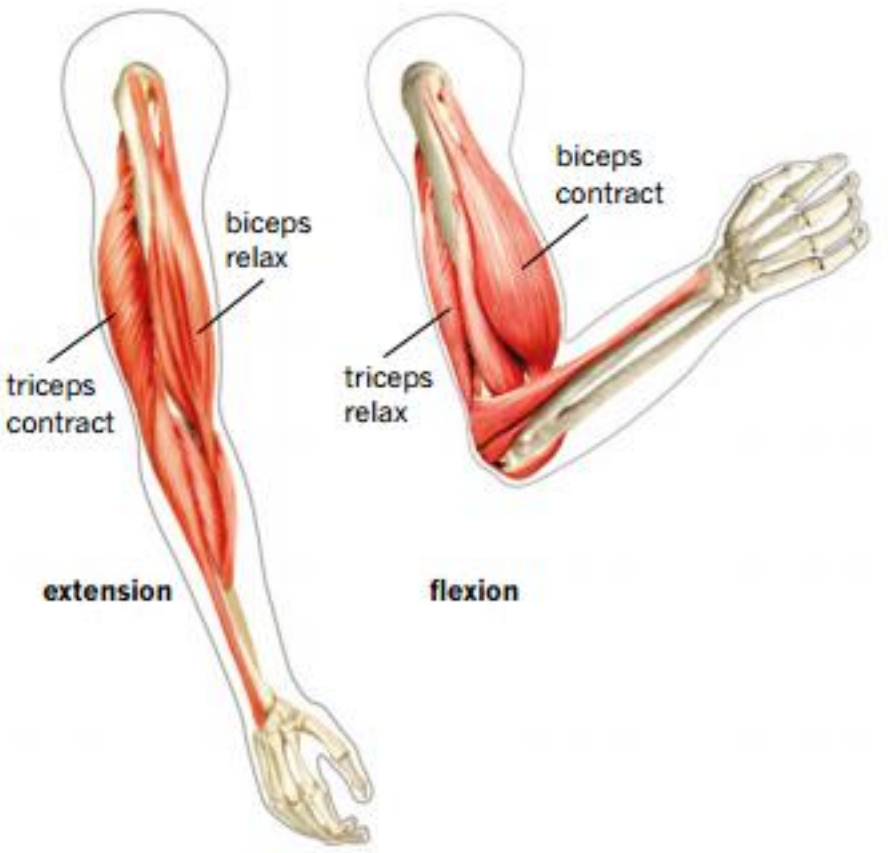


FIGURE 6.2.10 Extension (straightening) of the arm involves the biceps muscle relaxing and the triceps muscle contracting. The opposite action, flexion (bending) of the arm at the elbow joint, involves the biceps muscle contracting, while the triceps muscle relaxes.

Many reflexes are integrated with other parts of the body. For example, there is a withdrawal reflex of the leg similar to the withdrawal reflex of the hand in response to pain (Figure 6.2.8). However, if you tread on a nail, you cannot simply withdraw your foot; if you did, you would fall over. You must first shift your weight onto the other foot. In this situation, interneurons also carry instructions to the muscles of the opposite leg, making it brace to take your weight as you lift the painful foot. At a more complex level, increased numbers of interneurons in the nervous system (primarily in the brain of the CNS) provide more opportunity for synapses between neurons, thus allowing increased integration of information received from all parts of the body.

Processing stimuli

After a stimulus has been detected by a receptor, a message is sent along the axon of the receptor cell to the processing centre of the nervous system. In complex animals, this processing centre is made up of the brain and spinal cord of the CNS, as shown in Figure 6.2.11.

Different regions of the brain are associated with particular functions.

- The cerebral cortex (cerebrum) has areas associated with motor activity, sensory input, speech, sight and hearing.
- The hypothalamus receives information relating to the wellbeing of the body, and functions in maintaining homeostasis.
- The cerebellum is involved in the coordination of muscular activity, including posture, balance and movement.
- The brainstem has centres associated with the control of the heart, blood vessels and lung ventilation.

As well as coordinating information from all parts of the body to produce appropriate responses, the brain stores information so that responses can incorporate past experiences and learned information (memories).

In addition to the CNS, complex animals have a peripheral nervous system (Figure 6.2.12). The **peripheral nervous system (PNS)** includes sensory nerves, which carry information towards the CNS, and motor nerves, which carry signals to effector organs such as muscles and glands. The motor component of the PNS can also be subdivided into somatic (voluntary) and autonomic (involuntary) systems. The voluntary nervous system involves functions over which you have voluntary control, such as movement of the body by skeletal muscles.

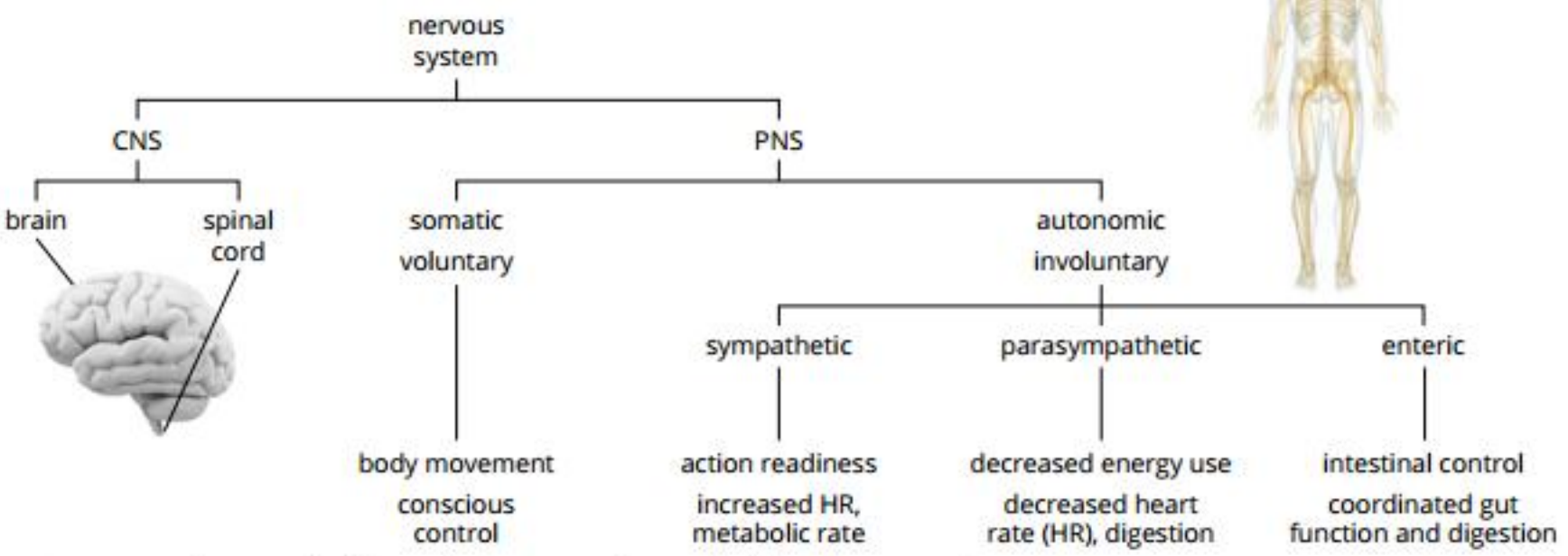


FIGURE 6.2.12 Components of the central nervous system and peripheral nervous system

The **autonomic nervous system**, shown in Figure 6.2.12, includes nerves involved in unconscious responses, such as constriction of pupils, secretion from glands and heart rate changes. It conveys signals to smooth muscle (internal organs), heart muscle and glandular tissues, and regulates the activities of the digestive, cardiovascular, excretory, respiratory and endocrine systems. Table 6.2.1 outlines the roles of the autonomic nervous system.

The correct balance of activity in different divisions of the nervous system, along with their interactions with the endocrine system, is central to maintaining homeostasis.

TABLE 6.2.1 Roles of the autonomic nervous system

System/role	Function/description
cardiovascular system	controls the rate and strength of the heartbeat and the distribution of blood to different organs by changing the diameters of arteries
digestive system	controls mixing, breakdown, movement and absorption of food through the gut and secretion of digestive enzymes in various regions of the gut
respiratory system	controls the diameter of major airways of lungs, rate of breathing, and the secretion of mucus over respiratory surfaces
excretory system	promotes emptying of the bladder and controls rate of production of urine by the kidneys
reproductive system	controls contraction of various parts of the reproductive tract in males and females, and thus the passage of eggs, sperm and embryos
metabolic regulation	controls the formation and release of hormones affecting overall metabolism
temperature regulation	controls cutaneous blood flow and sweating
eye function	controls the diameter of the pupil to regulate incoming light, secretion of tears and focusing of the lens

6.2 Review

SUMMARY

- The nervous system involves a more direct pathway of communication than hormonal responses. Control by nerves is generally rapid, short in duration, and precisely located.
- The neuron is the functional unit of the nervous system, and its basic structure and function are very similar across all groups of animals.
- Neurons are composed of different functional components including dendrites, soma (cell body), axon, myelin sheath, nodes of Ranvier, axon terminal and synapse.
- There are three main types of neurons: sensory neurons, interneurons and motor neurons.
- Neurons are excitable cells. The three basic steps involved in their function are:
  - generation of an action potential
  - conduction of the action potential along axons
  - chemical transmission to another cell across a synapse.
- Nervous responses require more energy than hormonal responses.
- Neuron signal transduction involves the conversion of the electrical impulse of the action potential into chemical signalling molecules, called neurotransmitters, and back again at the synapse between cells.
- Highly organised nervous systems have a central coordinating group of neurons (CNS—brain and spinal cord), and peripheral pathways of sensory and motor nerve fibres (PNS).
- A rapid unconscious nervous response is called a reflex.
- The simplest type of nervous response in a mammal, a monosynaptic reflex, involves just two cells.
- More complex circuits that include interneurons enable more control.
- The brain has four main regions which all have specific functions: cerebral cortex, hypothalamus, cerebellum and brain stem.
- Sensory receptors, often grouped into sense organs, monitor a wide range of conditions in an animal's internal and external environment and provide information that enhances the animal's ability to survive and reproduce.

KEY QUESTIONS

Describe

1 Explain what causes the release of neurotransmitters into the synaptic cleft.

2 Describe the function of the myelin sheath surrounding an axon.

3 Describe how a nerve impulse travels along an axon.

4 Identify the difference between a sensory neuron, motor neuron and interneuron.

Apply

5 Explain the role of interneurons in a nervous system response.

6 Explain the passage of a nerve impulse from the axon terminal to the postsynaptic neuron.

7 Complete the missing sections within the table below, noting points of difference between the structure and function of neurons.

	sensory neuron	**interneuron**	**motor neuron**
structure		short dendrites, long or short axon	
location	dendrites outside the spinal cord	within the CNS	dendrites and cell body in the spinal cord, axon outside the spinal cord
function		connects neurones	

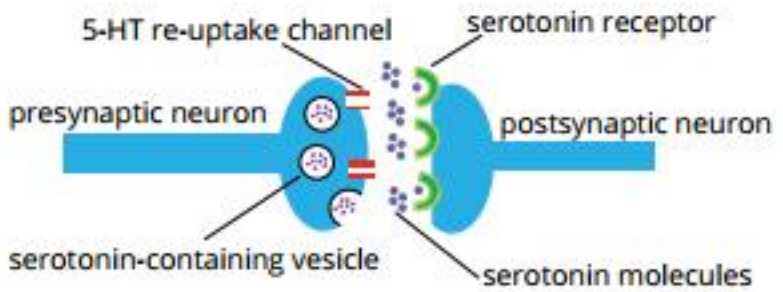
8 Represent, by drawing a flow chart, a reflex response for pulling your hand away from a hot frying pan. Label the neurons involved.

Analyse

9 Serotonin is an important neurotransmitter in the brain, which influences mood. Depression has been linked to serotonin imbalances in the brain. It has been postulated that depression is caused by low levels of serotonin in the synapse.

In the brain, serotonin is released from the presynaptic membrane. It diffuses across the synapse and attaches to receptors on the membrane of the next neuron. This stimulation results in an improved mood.

Unlike some neurotransmitters, which are broken down by enzymes in the synapse, serotonin is recycled back into the presynaptic neuron through special receptors called 5-HT re-uptake channels (or receptors) as shown below.



A group of drugs called selective serotonin re-uptake inhibitors (SSRIs) are one of the major treatments for clinical depression. The drugs work by increasing the amount of serotonin available in the synapse.

a Deduce what intended effect selective serotonin re-uptake inhibitors will have on action potentials in the postsynaptic neuron.

b Deduce how selective serotonin re-uptake inhibitors might decrease the re-uptake of serotonin.

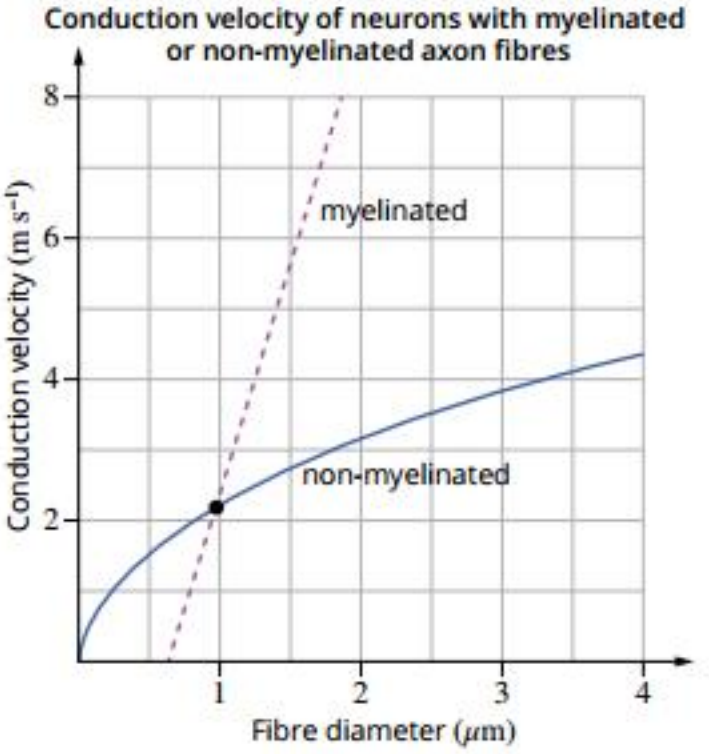
continued over page

6.2 Review *continued*

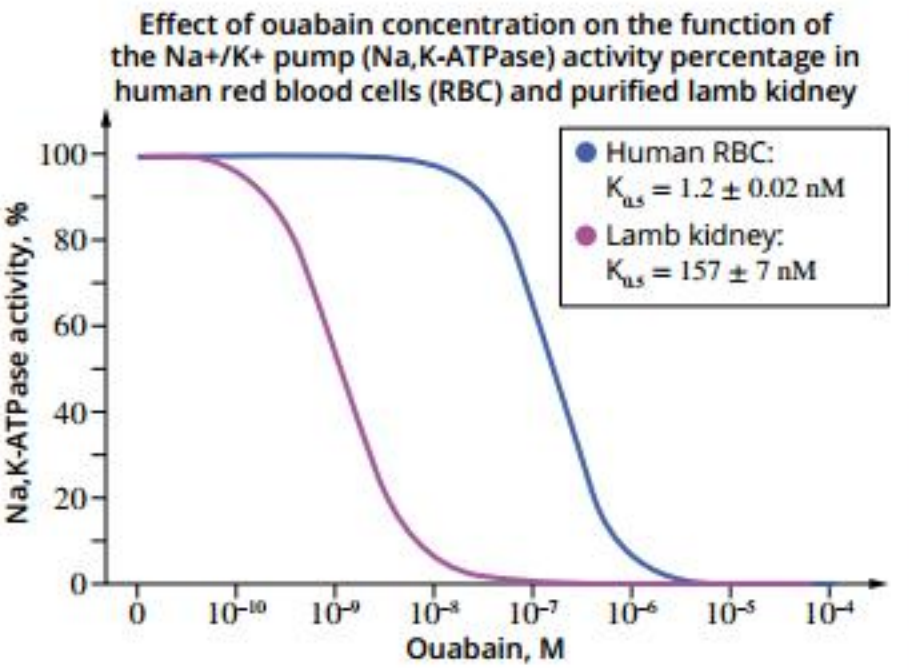
10 Not all axons have myelinated sheaths. The conduction velocity of a neural impulse depends on both the diameter of the axon fibre and whether or not it is myelinated or non-myelinated. The graph shows data on the conduction velocity (m s^{-1}) of neurons that have either myelinated or non-myelinated axon fibres, as collated and graphed by Rushton (1951) from data also published by Hursh (1939). The small circle on the non-myelinated curve marks the fastest recorded velocity (2.3 m s^{-1}) for an impulse travelling on the largest (1.1 µm) non-myelinated fibres of the sympathetic nervous system.

a Determine at what diameter of axon fibre the myelinated sheathed axon fibre begins to conduct an action potential at a greater rate than a non-myelinated fibre.

b The optic nerve in humans has a diameter of 3 µm and a conduction velocity of approximately 16 m s^{-1}. According to Rushton's findings, deduce whether the optic nerve has myelinated or non-myelinated fibres.



11 The graph below shows the effect of increasing ouabain concentration (M) on the function of the Na^+/K^+ pump (Na^+K^+-ATPase) activity % in human red blood cells (RBC) and purified lamb kidney.



a Outline the relationship between Na^+/K^+ pump (also called Na^+K^+-ATPase) function and ouabain concentration.

b Predict the ouabain concentration at which Na^+/K^+ (also called Na^+K^+-ATPase) activity would be 25% in human RBC.

c Explain why ouabain might be able to kill large animals, giving reference to the process of action potential in transmitting nerve impulses.

6.3 Hormonal control pathways

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- describe how hormones relay messages to cells displaying specific receptors via the circulatory or lymphatic system
- explain how receptor binding alters cellular activity, recognising that a cell's sensitivity to a specific hormone is directly related to the number of receptors it displays for that hormone.

Hormones help to control many of the functions within a living organism. Most hormones are used in negative feedback loops in conjunction with the nervous system, and so maintain a state of homeostatic equilibrium.

The production of hormones from the brain is initially stimulated by the nervous system. Hormones, in turn, can greatly affect the central nervous system of the organism, resulting in changed behaviour and development of the organism.

CHEMICAL MESSENGERS

A **hormone** is a signalling molecule produced in tiny amounts that can have relatively long-lasting effects on target cells. Hormones help regulate the growth and activity of cells in most animals and plants. A given hormone can only act on those target cells that have the hormone's specific receptor proteins. Non-target cells lack the specific receptor proteins and so remain unaffected.

A hormone produces its effect by altering the activity of the target cell or tissue. It can change cell membrane permeability or membrane potential by opening or closing ion channels. It may stimulate the production of enzymes or other proteins within a cell or can even activate or deactivate particular enzymes. Hormones can induce secretory activity or stimulate mitosis of a cell.

A minute amount of hormone can have a cascade effect, or a step-by-step amplification effect on the production of a large amount of a secreted product, final product or physiological response within the organism.

Animal hormones

In vertebrates, the endocrine system is responsible for coordinating many body functions, including growth, metabolism and reproduction. The endocrine system is made up of many glands and organs within the body that, along with some specialised tissues, synthesise and secrete hormones into the bloodstream (or in some cases the lymphatic system). The circulatory system transports the hormones from their site of production to the target cells and tissues. The main glands and organs of the human endocrine system are shown in Figure 6.3.1.

The production of hormones from the brain is initially stimulated by the nervous system. Hormones, in turn, can greatly affect the CNS of the organism resulting in changed behaviour and development of the organism.

Hormones can be broadly grouped into three main classes.

- Lipid hormones are a class of hydrophobic signalling molecules derived from fatty acids (**eicosanoids**) or cholesterol (**steroids**). Eicosanoids include prostaglandins, which are involved in cell growth, fever and inflammation. Steroid hormones help to regulate metabolism, salt and water balance, inflammation and sexual function. Examples of steroid hormones are testosterone, progesterone, oestrogen and **cortisol**.
- Peptide and protein hormones are a class of hydrophilic signalling molecules. An example of a peptide hormone is insulin, and an example of a protein hormone is growth hormone.

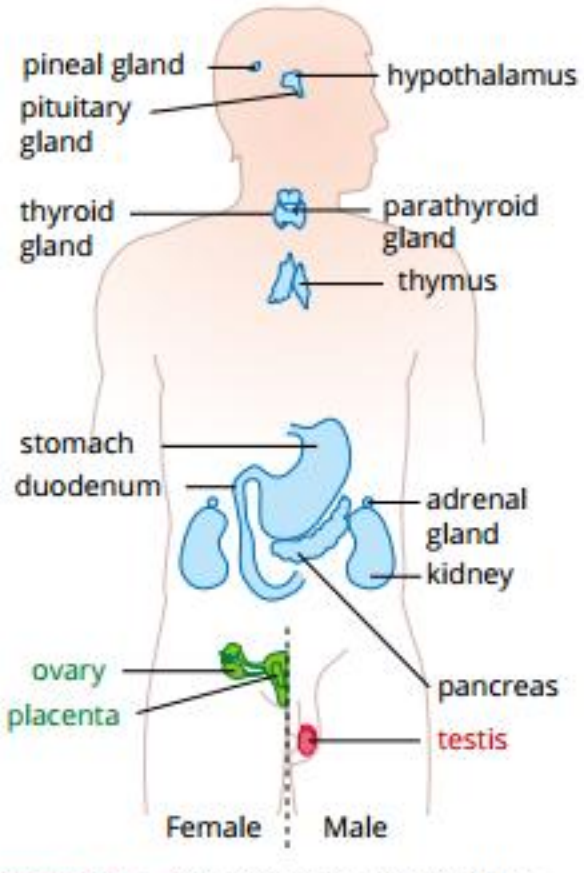


FIGURE 6.3.1 The organs and glands of the human endocrine system produce and secrete hormones into the bloodstream.

- Amino-acid-derived hormones are a class of small signalling molecules derived from the amino acids: tyrosine and tryptophan. They can be further divided into **catecholamines** and **thyroid hormones**. Thyroid hormones such as thyroxine are hydrophobic. Catecholamines are hydrophilic; examples are adrenaline and **dopamine**. Dopamine acts as both a neurotransmitter and a hormone.

A single hormone can trigger different responses in multiple target cells at the same time. For example, **adrenaline**, a hormone that is released from the adrenal glands, targets cardiac muscle cells, vascular smooth muscle cells and the various glands and organs of the digestive system. An increase of adrenaline in the bloodstream will result in an increase in heart rate and blood pressure and will simultaneously decrease digestive functions, preparing for a 'fight or flight' response.

Some common mammalian hormones, their sources, target tissues and functions are listed in Table 6.3.1.

TABLE 6.3.1 Common mammalian hormones, their sources, target tissues and functions

Gland (source)	Hormone(s)	Hormone class	Hydrophobic or hydrophilic	Target	Function
adrenal cortex	glucocorticoids	steroid	hydrophobic	many cell types	regulate glucose metabolism and stimulate fat breakdown
	mineralocorticoids	steroid	hydrophobic	kidney tubule cells	regulate reabsorption of salts
	androgens	steroid	hydrophobic	male and female reproductive organs, central nervous system, cardiovascular system, bones	stimulate puberty, regulate reproductive functions, support muscle development and bone density
hypothalamus	dopamine	amino-acid-derived	hydrophilic	anterior pituitary	inhibits release of prolactin
	growth hormone releasing hormone (GHRH)	peptide	hydrophilic	somatotroph cells in the pituitary gland	stimulates release of growth hormone
anterior pituitary	adrenocorticotrophic hormone (ACTH)	peptide	hydrophilic	adrenal cortex	promotes release of adrenal cortex hormones
	growth hormone (GH)	protein	hydrophilic	bone, muscle	promotes protein synthesis and growth
	follicle stimulating hormone (FSH)	protein	hydrophilic	ovaries	promotes development of follicle and secretion of oestrogen
	luteinising hormone (LH)	protein	hydrophilic	ovaries	promotes ovulation, development of corpus luteum and secretion of progesterone
	prolactin	protein	hydrophilic	mammary glands	stimulates milk synthesis and secretion
	thyroid stimulating hormone (TSH)	protein	hydrophilic	thyroid	promotes production and release of thyroxine
pancreas	insulin	peptide	hydrophilic	most cells	regulates blood glucose levels
thyroid	thyroxine	amino-acid-derived	hydrophobic	most cells	regulates cellular metabolic rate

Pituitary gland

The pituitary gland is located at the base of the brain, just above the roof of the mouth. In humans, it is about 1 cm in diameter and weighs about 0.5 g. Despite its relatively small size, the pituitary gland is often called the 'master gland' of the endocrine system because it produces many of the body's hormones, a number of which help regulate the production of other hormones around the body. Hormones secreted by the pituitary gland are also involved in a range of cellular processes including growth, reproduction, lactation, kidney function, skin pigmentation and regulation of the activity of the thyroid and the adrenal glands. Research has shown that the pituitary gland consists of two distinct parts, called the anterior and posterior pituitary gland.

Hypothalamus

Another endocrine gland, the hypothalamus, is located above the pituitary gland, and connects directly to the pituitary gland (Figure 6.3.2). The hypothalamus is responsible for detecting internal stimuli from all over the body and determining whether or not optimal conditions are being maintained. These internal stimuli trigger the production of releasing hormones from the hypothalamus. Releasing hormones are those that control and regulate specific hormone production in the pituitary gland. The combined functions of the hypothalamus and pituitary gland are vital for homeostasis.

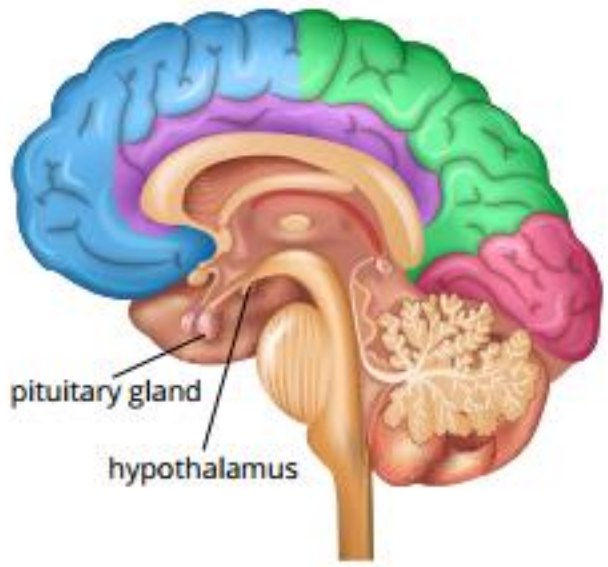


FIGURE 6.3.2 A cross-section of the human brain showing the location of the hypothalamus and pituitary gland

CELL SENSITIVITY AND RESPONSE

Number of receptors

The sensitivity of a cell to a specific hormone depends directly upon the number of receptors that that cell has in its membrane for that particular hormone. The greater the number of receptors for a particular hormone that a cell has, the greater degree of sensitivity that cell has to the hormone. If a cell, or structure, increases the number of receptors for a hormone, it is said to be upregulating. A subsequent decrease in the number of receptors is called downregulation and the sensitivity of that target cell to the hormone will be decreased. Additionally, the number of receptors that respond to a hormone can change over time, resulting in increased or decreased cell sensitivity.

Signal transduction

The process of converting the original stimulus signal into a response is called signal transduction (Figure 6.3.3). Signal transduction involves changing the form of the signal in some way. This may involve a change in the type of signalling molecule used, passing the signal into or out of a cell, or converting the type of signal from one form to another (for example, from a chemical to an electrical signal). The specific processes involved in the transduction of a particular signal depend on the signalling molecules involved.

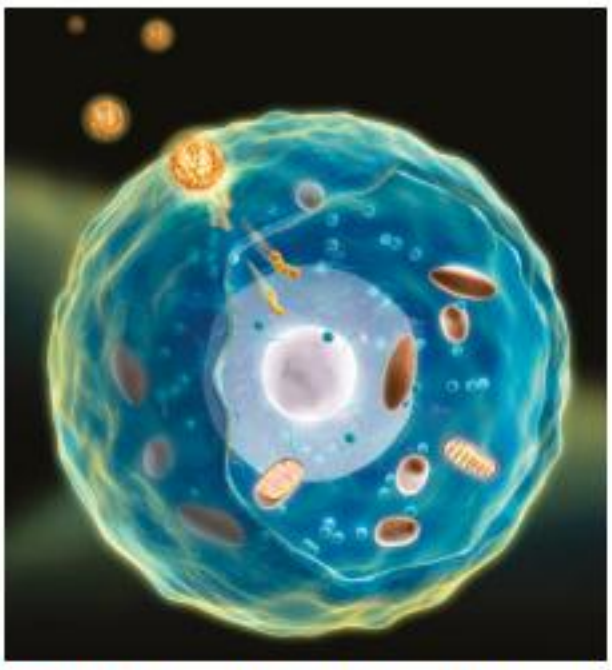


FIGURE 6.3.3 Signal transduction occurs when a signalling molecule from outside the cell (orange), activates a specific receptor located on the cell surface or inside the cell.

Stimulus–response model

The ability of a multicellular organism to detect and respond to stimuli relies on cells communicating with each other. As you learnt, cells communicate with each other through signalling molecules (hormones) (Figure 6.3.4).

The processes involved in a cell detecting and responding to a signalling molecule (hormone) are together known as signal transduction. The general characteristics of signal transduction depend on whether the signalling molecule is hydrophobic or hydrophilic.

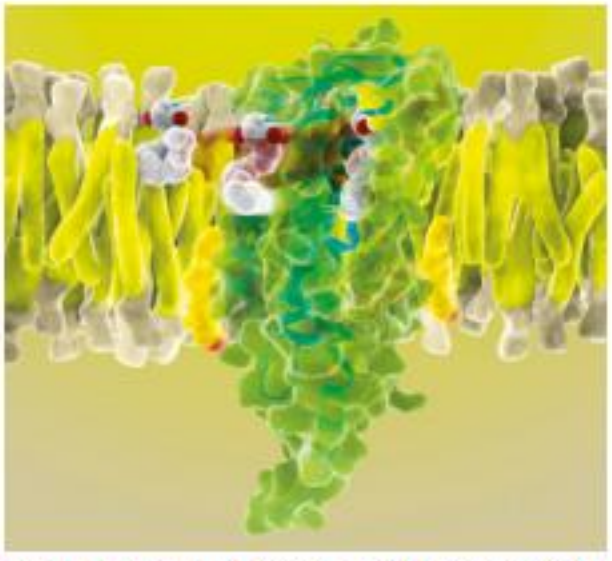


FIGURE 6.3.4 A digital illustration of a signalling molecule (mostly white) binding to a receptor (green)

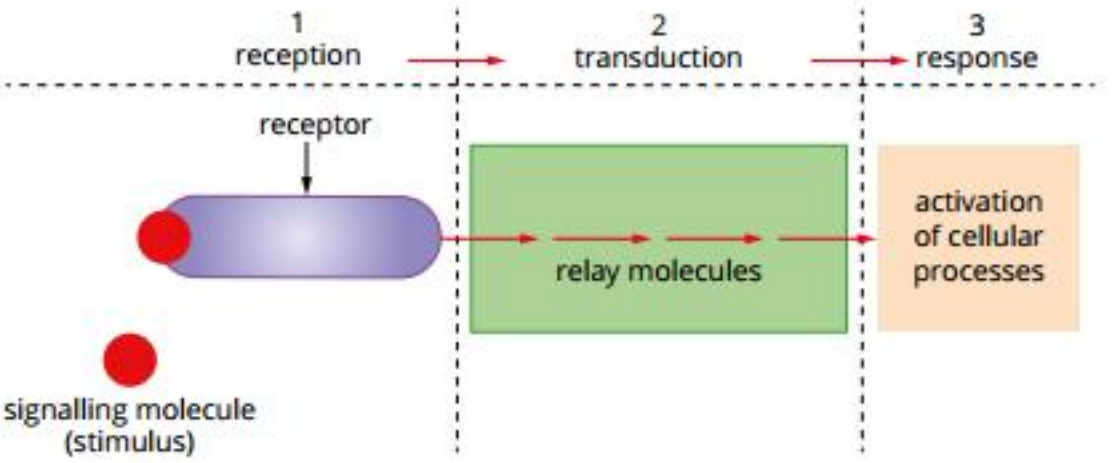


FIGURE 6.3.5 The stimulus–response model applied to the cell in terms of signal transduction

Signal transduction can be considered in terms of a stimulus–response model (Figure 6.3.5).

The stimulus–response model is a three-step process.

1 Reception—the detection of the signalling molecule (hormone) by a receptor.
2 Transduction—the relay of the signal into the cell.
3 Cellular response—the activation of a cellular activity or process.

Reception

Reception involves the detection of a signalling molecule (hormone) by a cell. The receptor that detects a signalling molecule (hormone) can be located on the surface of the cell membrane or in the cytosol or nucleus of the cell. The position of the receptor depends on whether the signalling molecule (hormone) is hydrophobic or hydrophilic. Receptors are specific and will only bind to particular signalling molecules (hormone) (Figure 6.3.6).

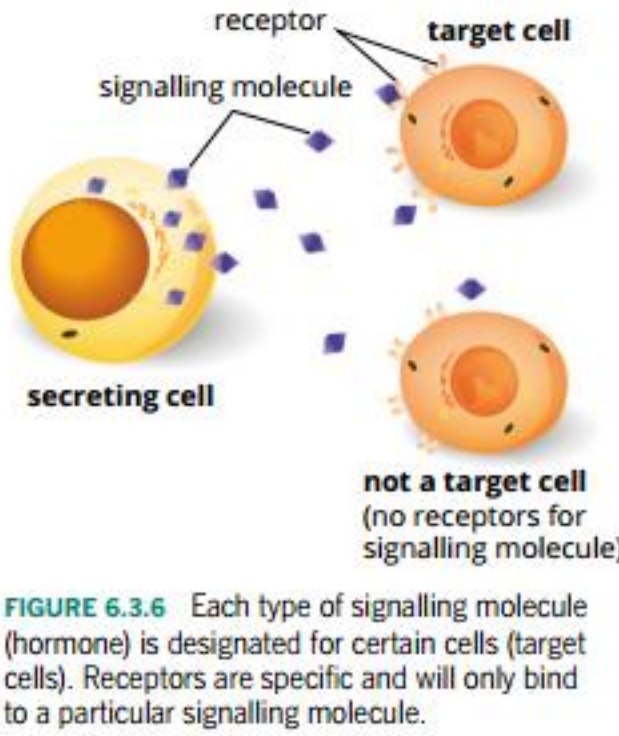


FIGURE 6.3.6 Each type of signalling molecule (hormone) is designated for certain cells (target cells). Receptors are specific and will only bind to a particular signalling molecule.

Not all cells are responsive to all signalling molecules. If a cell does not express the gene for a specific receptor, it will not have the receptors and hence that cell cannot respond to the complementary signalling molecule. For example, in the brain, neurons in certain neural pathways may express a defined set of receptors to be responsive only to the specific neurotransmitters used in that pathway.

Transduction

Transduction involves converting the signal into a form that can be relayed to reach its final destination within the cell and bring about a cellular response. Transduction may involve a one-step process in which a signalling molecule binds to one receptor and this complex produces a response. Alternatively, it may involve a multi-step process in which a signalling molecule binds to its receptor, leading to the sequential activation of different molecules in a chain of events. These multi-step transduction pathways are commonly called cascades.

Cellular response

Following transduction, a response is initiated. Cellular responses include any cellular activity such as gene transcription, the activation of enzymes or the secretion of signalling molecules by the cell. Responses can occur in the:

- nucleus
- cytosol
- cell membrane.

Signal transduction of hydrophobic signalling molecules

Hydrophobic signalling molecules are usually lipid-based molecules involved in gene regulation. Lipid-based molecules are lipid soluble; they can easily diffuse through the cell membrane. Inside the target cell, they bind to an intracellular receptor, either in the cytosol or in the nucleus.

Reception

Steroid hormones are examples of lipid-based hydrophobic signalling molecules that bind to receptors in the cytosol. Figure 6.3.7 shows some examples of steroid hormones. Once bound, the signalling molecule–receptor complex moves from the cytosol through nuclear pores to its final destination in the nucleus (Figure 6.3.8).

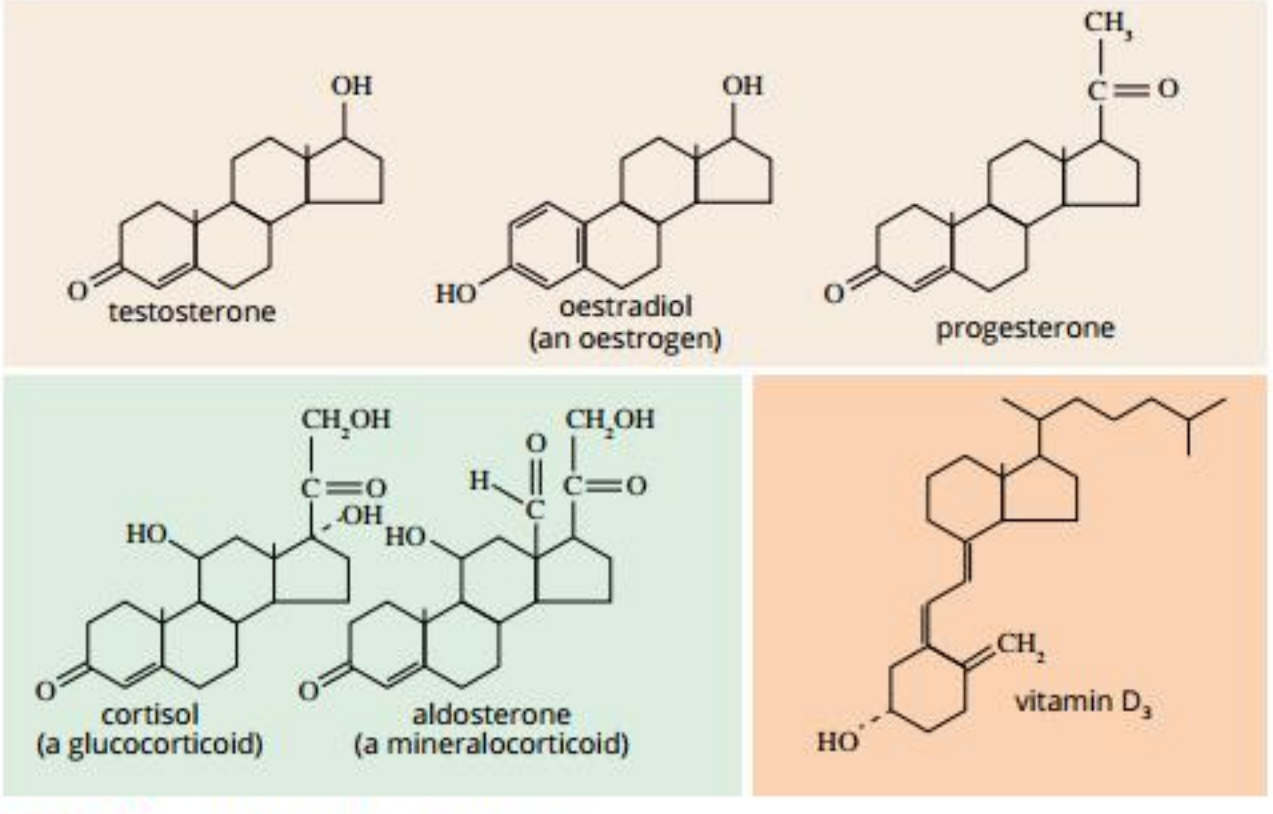


FIGURE 6.3.7 Examples of steroid hormones

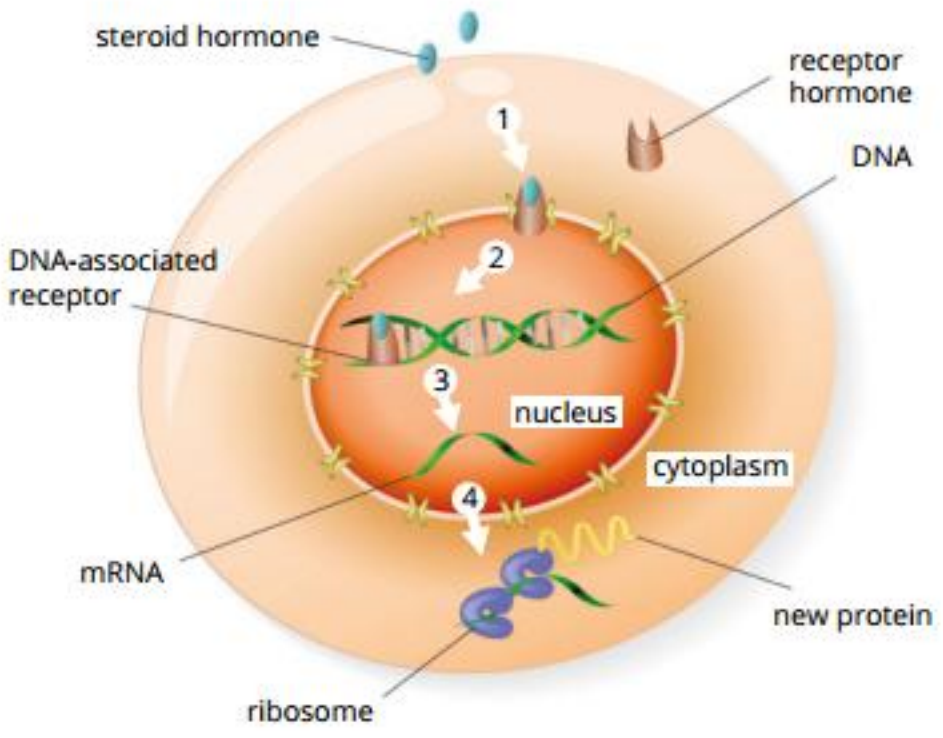


FIGURE 6.3.8 Once steroids are in the cytoplasm (1), they bind to receptors in the cytosol, then travel to the nucleus and move through pores in the nuclear membrane (2), which activates gene transcription (3) and causes proteins to be manufactured on the ribosome (4).

Transduction

In the nucleus, the signalling molecule–receptor complex acts as a transcription factor. Transcription factors are a group of proteins that help to initiate or regulate gene expression by either inducing or repressing gene transcription (e.g. copying a strand of mRNA from DNA). Figure 6.3.9 shows a molecular model of an androgen–receptor complex (signalling molecule–receptor complex) acting as a transcription factor and binding to DNA. The androgen receptor binds to hydrophobic signalling molecules called androgens, which are steroid hormones, such as testosterone. The binding of the androgen–receptor complex switches on the genes involved in development of reproductive organs and secondary sexual characteristics.

FIGURE 6.3.9 A molecular model of the DNA (red and blue) binding region of the androgen receptor (yellow and pink)

For example, testosterone is secreted by the cells of the testes in males. Testosterone travels in the bloodstream and enters all cells in the body. Only cells that contain androgen receptor molecules in the cytosol respond. In these cells, testosterone binds to the androgen receptor and activates it. The active form of the receptor then enters the nucleus and turns on specific genes that control male sex characteristics by binding onto a specific sequence of DNA known as hormone response elements.

Although traditionally thought of as male hormones, androgens are sex hormones produced in the testes and adrenal glands in men and ovaries and adrenal glands in women.

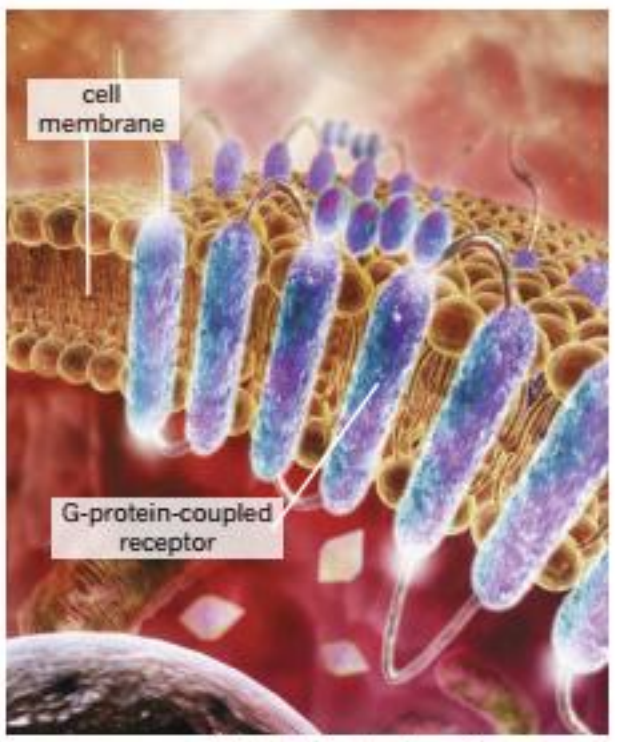


FIGURE 6.3.10 A digital illustration of a transmembrane protein (purple) in a cell membrane

Signal transduction of hydrophilic signalling molecules

Hydrophilic signalling molecules include a wide range of hydrophilic peptide hormones, neurotransmitters and cytokines. They are water-soluble and so are unable to diffuse through cell membranes.

Reception

The first step for all hydrophilic signalling molecules is to interact with receptors on the external surface of the cell membrane. The receptors for these signalling molecules are transmembrane proteins, which are made up of one or more protein molecules and span both layers of the cell membrane. These receptors are activated by the binding of molecules on the surface of the cell to induce cellular responses inside the cell (Figure 6.3.10).

Transduction

Transmembrane receptors have an extracellular domain that acts as a binding site for the signalling molecule and an intracellular domain that transfers the signal into the cell (Figure 6.3.11). When the signalling molecule binds to the extracellular part, the intracellular part of the receptor typically changes shape. This conformational change (a change in shape of a macromolecule) of the receptor results in the activation of molecules inside the cell (cellular responses).

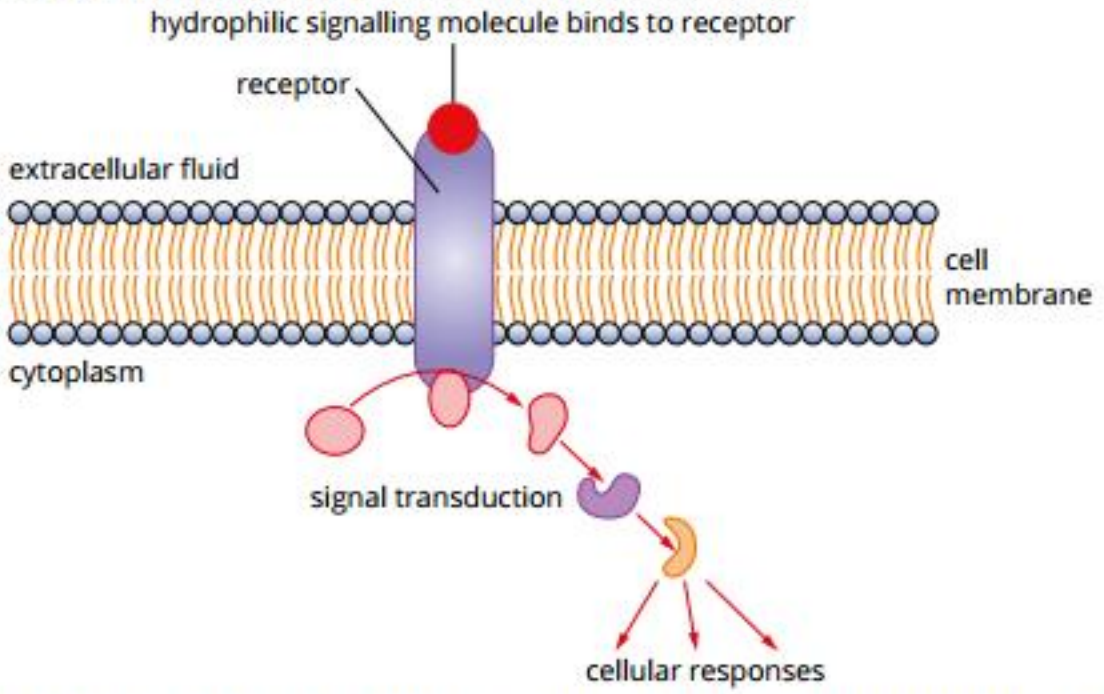


FIGURE 6.3.11 Hydrophilic signalling molecules interact with a specific membrane-bound receptor, causing the intracellular domain to change shape.

Transduction cascade

Transduction cascades involve a series of events in which a change in one molecule causes a change in another, which in turn causes a change in yet another, and so on. The molecules may be enzymes, channel proteins or cell structure proteins. For example, when hydrophilic signalling molecules, such as peptide hormones, activate metabolic pathways, a second messenger might be produced that activates a series of enzymes in a set order, so that the activation of one enzyme causes the activation of the next in a sequence or cascade (Figure 6.3.12). At the end of the cascade, molecules are activated that bring about a cellular response.

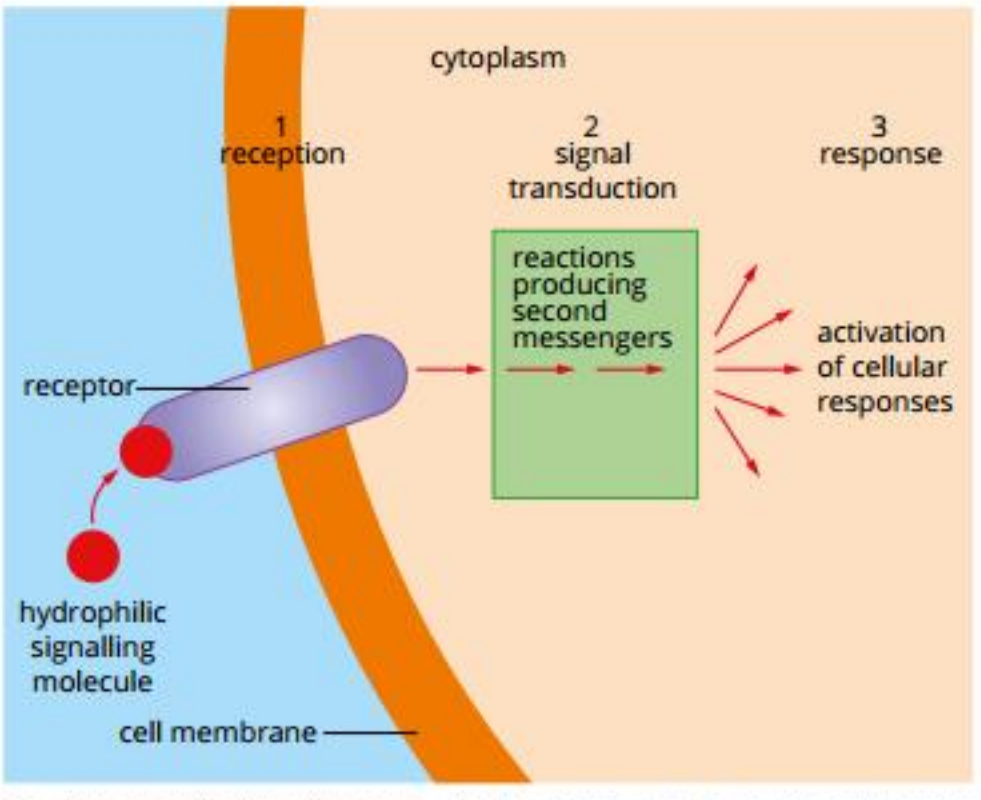


FIGURE 6.3.12 A hydrophilic signalling molecule binds to the receptor on the cell membrane, leading to a series of reactions that activate a cascade of enzymes or other molecules in the cell, leading to a range of possible cellular responses.

Cellular response

Gene regulation is vital for a cell to adapt and respond to incoming stimuli. The cell's need for enzymes and products such as signalling molecules fluctuates.

You have already learnt that hydrophobic hormones are able to cross the cell membrane. Once within the cytosol or nucleus, they form a complex with their receptor and in the nucleus this complex acts as a transcription factor.

Some hydrophilic hormones elicit a similar response but through a different transduction process. Peptide and protein hormones can activate protein receptors in the cell membrane that, in turn, activate a signal transduction cascade. The cascade ends with a functional transcription factor, leading to gene regulation (Figure 6.3.13).

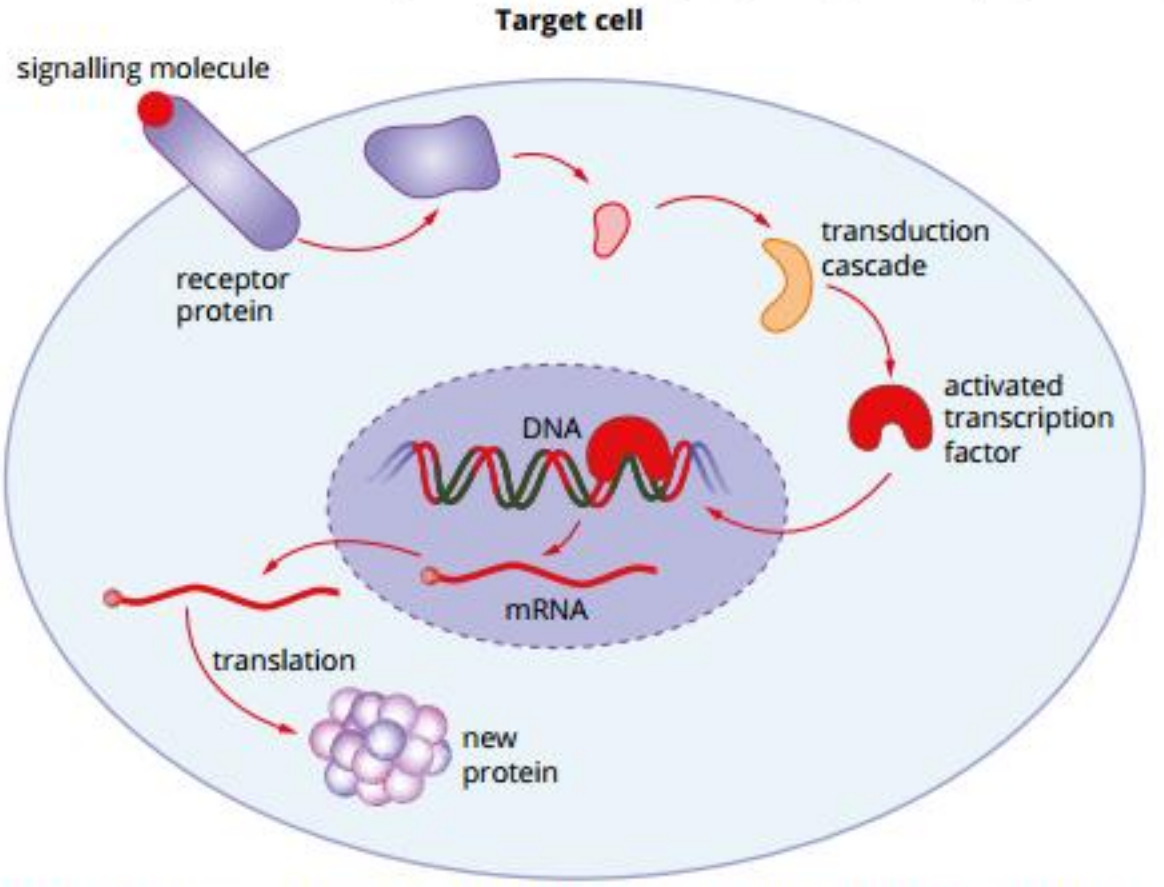


FIGURE 6.3.13 A hydrophilic signalling molecule interacts with its membrane-bound receptor, causing transduction of the signal into the cell with the resultant gene expression.

Signal transduction can result in the inhibition or activation of enzymes in the cytosol of the target cell. Enzymes regulate cellular processes by catalysing chemical reactions, so inhibiting or activating enzymes will decrease or increase cellular functions respectively.

Two distinct fluids move through the human body: blood travelling via the circulatory system and lymph moving through the lymphatic system. Both systems are able to transport hormones to specific target cell receptors in the body.

Blood carries oxygen and nutrients to the body's cells, and carries waste materials away, but also carries various hormones. One example of a hormone transported via the circulatory system is insulin. When glucose levels in the blood are high, insulin is released from the pancreas. Insulin binds to insulin receptors on the liver cell surface, which allows the receptor to activate second messenger molecules and initiate multiple transduction cascades. In liver cells, the pathway initiated by insulin leads to the activation of enzymes for glycogen synthesis, fatty acid synthesis, increased glycolysis and an increase in glucose transporters in the cell membrane for the uptake of glucose into the cytoplasm.

The lymphatic system (lymph, lymph nodes and lymph vessels) supports the circulatory system by draining excess fluids and proteins from tissues back into the bloodstream, thereby preventing tissue swelling. Lymph is a colourless fluid, yet unlike the circulatory system, which relies on a heart to pump blood around, the lymphatic system has no pump. Lymph is moved through the lymphatic system by the movement of muscles and the pulsing of arteries. As hormones travel in the blood circulation to reach their target sites, some hormones can also travel in the lymphatic system where they are destroyed in the liver and removed by the kidneys.

6.3 Review

SUMMARY

- Multicellular organisms produce signalling molecules as a form of intercellular communication.
- Signalling molecules transmit information about an internal or external stimulus to other cells, including, ultimately, the effector cells, which enact the response.
- Signalling molecules can be classified according to their chemical structure, their source and their mode of transmission.
- Animal hormones:
  - are produced by organs and glands in animals
  - can be hydrophilic or hydrophobic.
- The stimulus–response model outlines the three main steps involved in signal transduction in cells:
  - reception—the detection of a signalling molecule (the stimulus) by its specific receptor (including the physical binding of the signalling molecule to the receptor)
  - transduction—the transformation of the signal in terms of form, type of signalling molecule and the passage into and out of a cell
  - response—the change in cellular activity as a result of the initial stimulus.
- Hormones are transported through blood via the circulatory system to a specific target cell. Hormones are also able to be transported to specific target cells via lymph in the lymphatic system.
- Cellular responses depend on the stimulus and the type of cell, the signalling molecules and response molecules involved.

KEY QUESTIONS

Describe

1 Describe the feature of a target cell that makes it receptive to a particular hormone.

2 Describe the terms reception, transduction and cellular response.

3 Identify the differences in the process of reception when comparing hydrophilic and hydrophobic signalling molecules.

4 Explain the terms 'upregulating' and 'downregulating' in relation to hormone receptors.

Apply

5 Hormones and neurotransmitters are both signalling molecules. Explain the difference between the two.

Analyse

6 Summarise the signalling process involved when a steroid hormone (such as testosterone) acts on a target cell.

7 Hormones can be broadly grouped into three main classes. Describe each of the classes and provide an example for each.

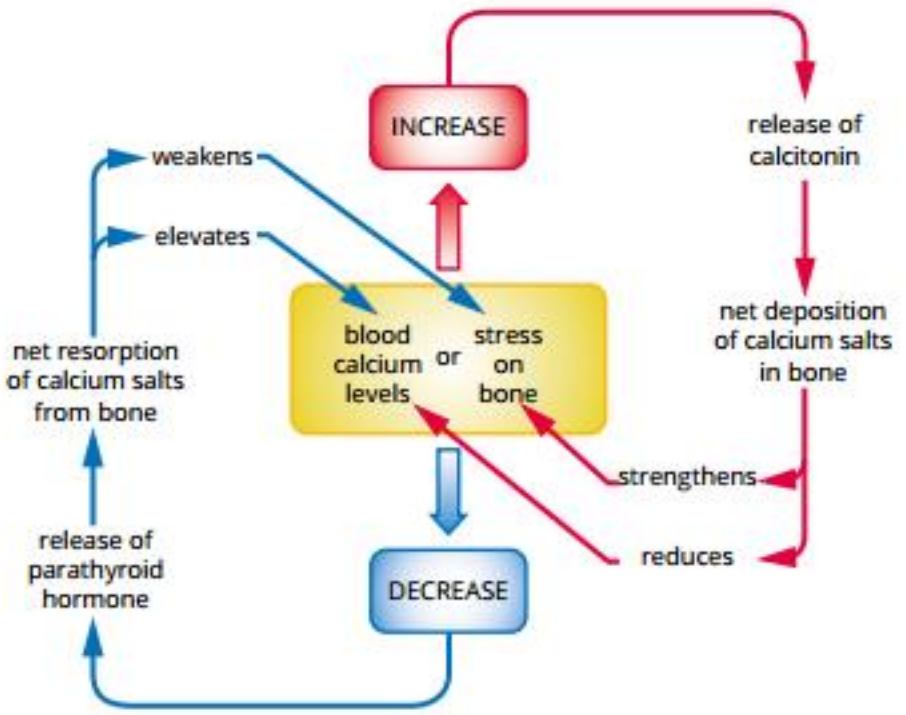
continued over page

6.3 Review *continued*

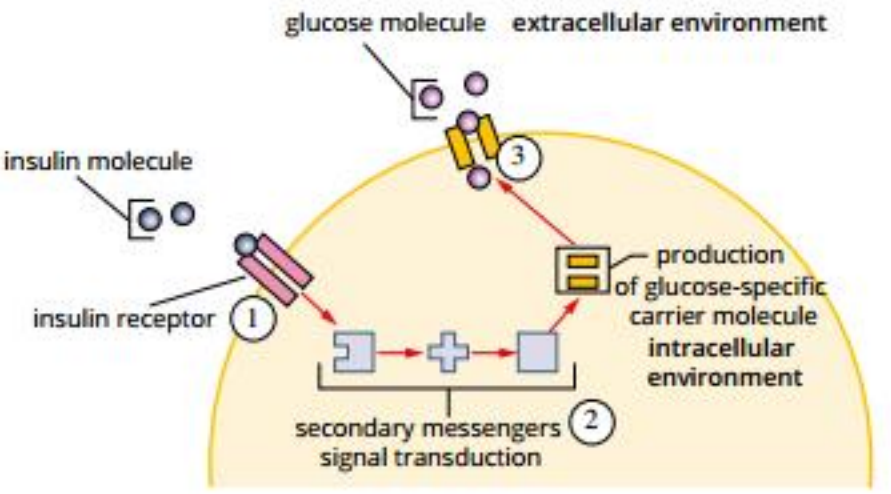
8 Many people in Victoria become vitamin D deficient through the winter months because of lack of sun exposure. In the absence of vitamin D, dietary calcium is not absorbed efficiently from the digestive tract.

a Use the diagram below to describe the likely effect that this will have on the relative concentrations of parathyroid hormone in the blood.

b Low levels of calcium can lead to problems at neuromuscular junctions, resulting in poor muscle control. Determine why this observation is valid.



9 The diagram below shows a summary of the steps in an insulin signalling pathway that results in increased glucose uptake by the human body. When levels of glucose are low, insulin binds to the insulin receptor to initiate a response.



a What type of molecule is insulin?

b Outline the three stages of cellular signalling as seen in the diagram above (1, 2 and 3).

c What type of receptor is the insulin receptor?

d Explain the response (location 3 in the diagram above) that occurs when insulin hormone binds to a target cell.

e What would occur if the number of insulin receptors on a target cell is increased?

6.4 Thermoregulation

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain thermoregulatory mechanisms of endotherms, including (1) structural features: insulation, brown adipose tissue and increased mitochondria in cells; (2) behavioural responses: kleptothermy, hibernation, aestivation and torpor; and (3) physiological mechanisms: evaporative heat loss, thermogenesis, vasodilation and vasoconstriction
- explain thermoregulation in humans, including the role of sweating, shivering, vasodilation and vasoconstriction using feedback control diagrams
- analyse feedback-control diagrams to identify the stimulus, receptors, control centre, effectors and communication pathways in different scenarios.

Organisms and cells are constantly experiencing changes in their environment. These changes to the internal and external conditions can adversely affect the survival, growth and function of the organism. The internal environment of an organism must always remain within tolerable limits, even when conditions in the external environment fluctuate widely. When a change occurs in the external environment, an adjustment must be made to the internal environment.

THERMOREGULATION IN ENDOTHERMS

Thermoregulation is when an organism regulates its body temperature either by increasing its metabolic activity or through behavioural responses. As you will recall from Chapter 3, all our biochemical mechanisms are driven by enzymes. Enzyme efficiency in catalysing chemical reactions depends on the organism maintaining an optimal internal temperature. Should thermoregulation fail because of an inability to cope with extremes of external temperatures, enzyme function decreases to a level unable to sustain life. But as you will learn in this module, some animals have adapted very well to living in extreme climatic conditions.

Homeostatic mechanisms

Regulation of body temperature in humans and other **endotherms** involves a complex negative feedback pathway with several sensory inputs and many effector responses that act together to maintain a stable body temperature. The control centre for measuring the body set point temperature in humans (37.2 ± 0.6°C) is in the hypothalamus. A change in the temperature of the hypothalamus initiates regulatory responses that can either reduce heat loss, or initiate heat production or heat exchange.

Endotherms (from the Greek *endon*, meaning 'within', and *thermos*, meaning 'heat') are warm-blooded animals that can generate heat to maintain their own body temperature.

Detecting temperature change

Regulation of temperature in humans is an example of the way different sensory receptors work together to produce an integrated response. Arterial blood has the most constant temperature. The relatively constant temperature of many other parts of the body indicates that they are well supplied with arterial blood.

In endotherms, a group of temperature-sensitive cells in the hypothalamus act as misalignment detectors, triggering homeostatic responses if blood temperature deviates from the optimal temperature range, or set point. Lowering or raising the temperature of the hypothalamus initiates regulatory changes in heat production or heat exchange.

Temperature receptors are also found in the skin. If these receptors detect a decrease in environmental temperature, they initiate regulatory responses, such as decreased blood flow to the skin to reduce heat loss, and behavioural changes, such as moving into a warmer or more sheltered environment. These responses take place long before there has been any change in the internal temperature of the body. Skin temperature receptors act as disturbance detectors, detecting changes in the external environment and triggering responses before there is a change in core body temperature.

As the environmental temperature falls, disturbance detectors stimulate responses that reduce heat loss and increase heat production. The reverse occurs as environmental temperature increases. If the arterial temperature falls despite the regulatory responses that have been initiated, or if it rises because the responses made have been too effective, these changes will be detected by the misalignment receptors in the brain, which will fine-tune the temperature-regulating mechanisms.

The function of the disturbance detectors in the skin is to reduce fluctuations in arterial blood temperature, providing a more precise control around the set point level than there would be if misalignment detectors (the brain's temperature receptors) alone were involved.

Heat loss

Organisms are constantly exchanging heat with their environment. This heat exchange occurs through four mechanisms (Figure 6.4.1):

- Conduction—occurs when the temperature of the organism and the environment are different. Heat exchange is a result of direct contact (e.g. a lizard basking on a warm rock).
- Convection—is the transmission of heat from a warmer region to a colder region, resulting from the movement of liquid or gas (e.g. heat moves from the inside of living organisms to the body surface by convection).
- Radiation—occurs all the time, without direct contact, regardless of temperature differences between the organism and their environment (e.g. heat radiating from dark coloured surfaces).
- Evaporation of water causes heat loss. This occurs most rapidly when the air is hot and dry (e.g. sweating).

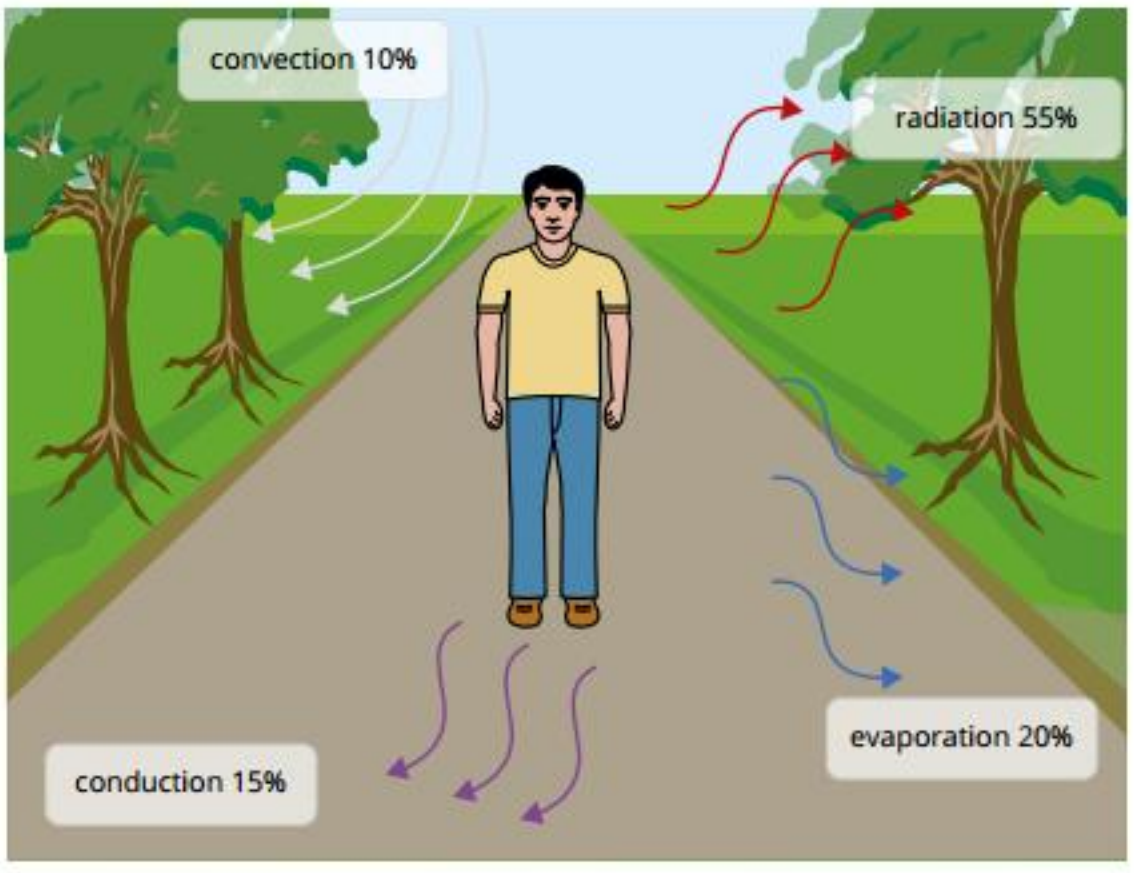


FIGURE 6.4.1 Methods of heat exchange with the environment and the average amount lost via each route in a human at rest

Structural features

All endotherms have structures that help them to conserve or to release heat. Examples are:

- special body coverings and blubber to insulate against cold
- vascular body parts such as ear size to reduce (small ears), or to increase (large ears), heat exchange with the environment
- brown adipose (fat) tissue
- increased numbers of mitochondria for heat generation.

Body coverings and insulation

The emperor penguin (*Aptenodytes forsteri*) has many structural features to cope with life in the harsh Antarctic climate. Penguins have four layers of thick, scale-like feathers, creating a windproof coat (Figure 6.4.2). They also have thick blubber to keep them warm while swimming in the icy ocean. Juvenile penguins have soft down for insulation, which is a more effective insulator on land than the adult feathers but of little use in the sea. Juveniles must moult before they can swim. Penguins and other animals in cold climates tend to have bodies with a small surface-area-to-volume ratio to assist them in conserving body heat.

Marine and seagoing mammals also have a thick layer of blubber beneath their skin. Polar bears and fur seals have a dense layer of fur that aids in insulation on land but need a layer of blubber for insulation while swimming. Depending on their size, whales have a very thick layer of blubber to conserve their body heat (Figure 6.4.3).

FIGURE 6.4.2 The emperor penguin (*Aptenodytes forsteri*) has mastered living in extremely cold environments. The thick layer of feathers and blubber are two of many important features that make this lifestyle possible.

FIGURE 6.4.3 These scientists are dissecting a dead male sperm whale (*Physeter macrocephalus*) found on Henne Strand, Jutland, Denmark. The whale was 14 metres long and weighed about 30 tonnes. Cutting tools and a backhoe loader were used to remove large portions of the whale's tissue. The thick layer of blubber is visible beneath the skin. The tissue samples and skeleton are used for further study by the scientists.

Vascular body parts

Features of animals living in hot, dry climates include large ears, long tails or a long body. When the extremities are highly vascular, it enables the animals to release heat efficiently and keep their bodies cool. The greater bilby (*Macrotis lagotis*) of the Australian inland, shown in Figure 6.4.4, is an example of a desert dweller with highly **vascularised tissue** (ears). Highly vascularised tissues contain many blood vessels.

This works in reverse with animals such as the Arctic fox (*Alopex lagopus*). Figure 6.4.5 shows their very small ears, which limit vascular heat loss.

FIGURE 6.4.4 The greater bilby (*Macrotis lagotis*) has highly vascularised ears that allow it to release heat rapidly, lowering the body temperature in the extreme desert heat.

FIGURE 6.4.5 The Arctic fox (*Alopex lagopus*) has very small ears that allow it to limit heat loss in freezing Arctic conditions

Brown adipose tissue

Many small animals with high metabolic rates undergo either hibernation or torpor during winter (see *Torpor* below). Their metabolic rate and body temperature drop down quite low during this time. Brown adipose tissue, or brown fat, is a structure that aids in the transition from hibernation to a full metabolic functioning, awakened state.

FIGURE 6.4.6 The endangered mountain pygmy possum (*Burramys parvus*) lives in Australia's alpine region. It is the only marsupial known to store food and hibernate for extended periods.

FIGURE 6.4.7 Emperor penguin chicks (*Aptenodytes forsteri*) huddle together for warmth.

FIGURE 6.4.8 Although a very good thermoregulator, the echidna (*Tachyglossus aculeatus*) readily enters torpor when food is scarce and temperatures are low.

Brown fat cells are packed with many large mitochondria (which contribute to its colour). This tissue has a rich blood supply and many sympathetic nerve endings. Unlike typical mitochondria that synthesise ATP, these mitochondria have been uncoupled from that mechanism, and focus on the oxidation of fat. The heat given off is picked up by the vascular system and distributed to the vital organs of the animal. The heat production is switched on by either adrenaline or nervous stimulation. Figure 6.4.6 shows a mountain pygmy possum (*Burramys parvus*), which has reserves of brown fat. It is the only marsupial known to hibernate for extended periods of time and is fully adapted to living solely in alpine regions of Australia.

Increased mitochondria in cells

ATP is produced primarily within the electron transfer chain of the mitochondria. Animals with high metabolic rates generally have a larger number of mitochondria per cell. This allows them to generate heat, although their higher rate requires them to eat more food than ectothermic animals (such as snakes and lizards).

Behavioural responses

In addition to physiological homeostatic mechanisms, animals use behavioural responses to protect themselves from, or to adjust to, their environment.

Kleptothermy

If you have ever huddled together with your friends on a cold and windy day, you have practised **kleptothermy**. Kleptothermy is the thermoregulatory behavioural response of sharing another organism's body heat.

Huddling is the prime example of this. Many animals use huddling to cope with cold temperatures. Figure 6.4.7 shows emperor penguin chicks, thousands of which can huddle together for warmth in the spring, when they begin to develop their adult plumage. By huddling, penguins decrease the surface area of the group exposed to the harsh environment. They continually change which birds are on the outside of the group, so that each takes a turn in the freezing cold winds.

Torpor

Torpor is a physiological state in which the metabolic rate is lowered to save energy. This enables an organism to cope with environmental stresses such as extreme cold or heat or decreased food or nutrient availability, and torpor can occur over short or long periods.

A long period of torpor is often called dormancy. Hibernation and aestivation are different forms of prolonged torpor.

Hibernation is prolonged torpor that occurs in winter. Over summer and autumn, the animal builds up a thick layer of brown body fat that will provide them with energy during the hibernation period in winter. During hibernation, the animal can decrease its metabolic rate so that its body temperature falls to almost that of its surroundings. Accordingly, its heart rate and oxygen consumption also become very low and very little energy is required to maintain life. When external temperatures fall below freezing, the animal will either die, or follow one of two options. Its metabolic rate will increase and the animal will awaken out of torpor. Alternatively, the animal's thermoregulatory system will increase its metabolic rate just enough to keep the animal's internal temperature at about 5°C. With such a low metabolic state, heat exchange with the external environment ceases to become an issue because the internal temperature of the animal is close to the external temperature (when above freezing).

An example of this is the echidna (*Tachyglossus aculeatus*) (Figure 6.4.8), which can maintain its normal body temperature even at freezing temperatures. However, if low temperatures are combined with a lack of food, the echidna readily becomes torpid. It regulates its body temperature at around 5.5°C, its heart rate decreases from an average of 70 beats per minute (bpm) to just 7 bpm, and its oxygen consumption becomes about one-tenth of its normal rate.

When in torpor, animals show little response to either noise or touch. Without the metabolic requirement of maintaining a high body temperature, an animal can live off its fat reserves for extended periods of time under unfavourable conditions. And while virtual suspended animation saves energy, arousal, or coming out of torpor, is an active process where the animal's metabolic rate increases to a maximum level, as does their oxygen consumption, until the animal is fully revived. In the case of small animals undergoing torpor for 10-hour periods, 75% of the energy consumption over that period is used in the arousal stage at the end.

Hibernation occurs mostly in mammals, but some species of birds also hibernate. Bears, bats and squirrels are examples of animals that hibernate.

Aestivation is prolonged torpor under hot and dry conditions. Examples of aestivating animals are snails, frogs, crocodiles, tortoises, lungfish and some birds.

Green-striped burrowing frogs (*Cyclorana alboguttata*) (Figure 6.4.9) inhabit semi-arid to arid regions of eastern Queensland and northern New South Wales. The frogs aestivate when there is a prolonged drought. These frogs spend up to 9 months of the year in aestivation. During this time, they live underground in small burrows and do not eat. Research has shown that during this time they can reduce their metabolic rate by up to 80%. This allows them to survive these underground periods just on their store of body fat. By burrowing, they have insulated themselves from the hot arid conditions outside and so do not experience heat stresses.

FIGURE 6.4.9 The green-striped burrowing frog (*Cyclorana alboguttata*) aestivates for up to nine months of the year underground.

Torpor is an example of both a behavioural response (retiring to a cave or seeking shelter and going to sleep) and a physiological mechanism (the slowing of the heart, breathing and metabolic rates associated with periods of torpor).

Other animal behaviours

Other animal behaviours that are considered adaptations in animals and which help them to survive extreme environmental conditions include seeking or leaving shade or shelter, and evaporative cooling to lower temperature. Adaptations are responses that aid homeostatic mechanisms and maintain health and survival.

Many desert animals have behavioural adaptations that are very important in regulating the rate of heat exchange with their environment. An example is the central netted dragon (*Ctenophorus nuchalis*), shown in Figure 6.4.10. To raise its body temperature, this lizard emerges from under a rock and basks in the sunshine, spreading itself out at right angles to the Sun's rays. To lower its body temperature or reduce the rate of increase in body temperature, the lizard orientates its body parallel to the Sun's rays, minimising the exposed surface area, or simply retreats beneath a rock or into a burrow.

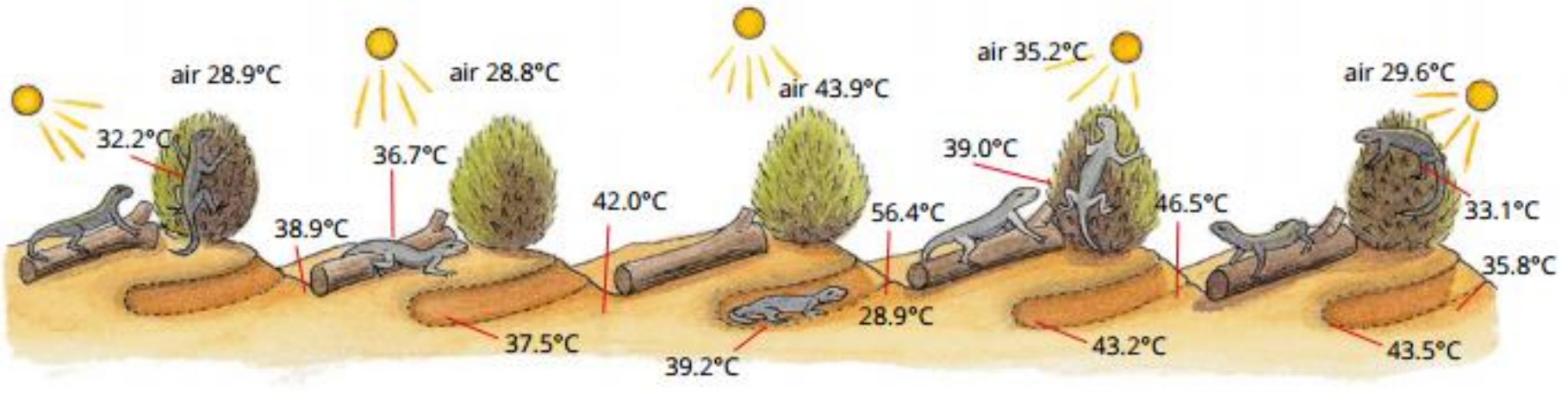


FIGURE 6.4.10 The central netted dragon (*Ctenophorus nuchalis*) has adapted its behaviour to desert conditions, regulating its temperature throughout the day by seeking shade or basking in sunlight.

Some animals such as desert snakes and tortoises adopt nocturnal behaviour during summer to prevent overheating. They move only in the cooler evening, avoiding the extreme heat of the day. Animals may also seek shelter to increase their body temperature when it is cold or windy.

Many land animals use evaporative cooling to lower their body temperature by releasing heat into the environment. Although it is a physiological mechanism it is achieved by behavioural responses, such as:

- panting or licking limbs
- spraying water on the body
- wallowing in mud or water
- mouth gaping
- gular fluttering (flapping membranes in the throat by birds to increase evaporation)
- urohidrosis.

Panting or licking limbs enable animals to release heat effectively using evaporative cooling. For example, kangaroos lick their paws, and animals such as dogs, gazelles and foxes, pant. The fennec fox (*Fenecus zerda*) has been observed panting at a rate of 690 times per minute after chasing prey. The rate of panting is proportional to the amount of air flowing over the tongue. If animals can flatten their tongue, increasing surface area, while increasing their panting rate, then the cooling effect is greater. Sometimes even penguins have to pant. In warmer weather, they also hold their flippers out of the water so that both surfaces are exposed and can release heat via evaporative cooling.

Spraying water on the body is commonly seen in elephants (Figure 6.4.11) but is also a behaviour used by many other animals.

FIGURE 6.4.11 A female African elephant (*Loxodonta africana*) splashes water over her body. She is using evaporative cooling to control her body temperature. Mud remaining on the elephant's skin provides protection against solar radiation.

Physiological mechanisms

Animals display an astounding complexity in the **heat exchange** system that they use. Heat exchange systems are physiological systems that allow the organism to rid themselves of heat, conserve heat, or selectively by-pass insulation layers. They do this through the use of **counter-current blood flow**, vascular switches and evaporative cooling.

Heat exchange for cooling

Heat exchange systems work in different ways in different animals. An example is the gemsbok oryx (*Oryx gazella*), which is a desert ungulate (hooved animal, Figure 6.4.12). It needs homeostatic mechanisms to counteract the heat in deserts, dehydration and the potential to die from overheating vital organs. Counteracting the heat is achieved through a network of small arteries that intertwine with a network of veins (the **carotid rete system**) to cool the blood before it enters the brain, as shown in Figure 6.4.12b.

The blood in the veins is cooler because it has experienced evaporative cooling in the nostrils. It passes in the opposite direction to the warmer blood from the body in the arteries and the heat flows from the hotter to the cooler blood. This process is known as counter current heat exchange and cools the blood entering the brain by several degrees.

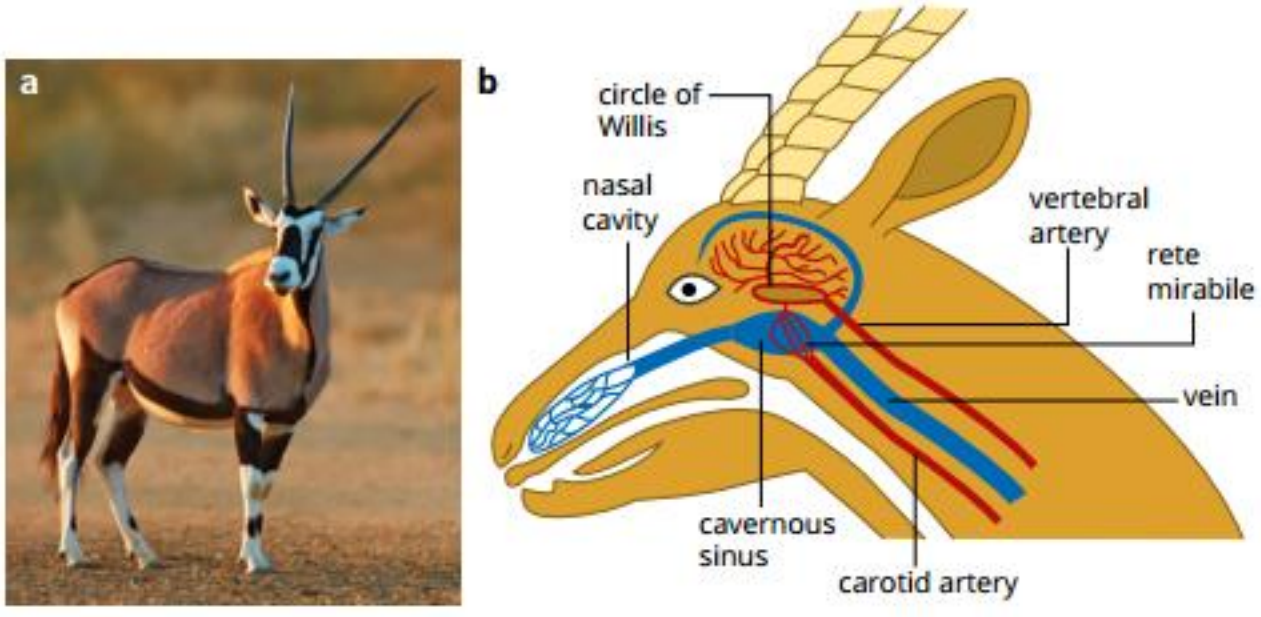


FIGURE 6.4.12 (a) A gemsbok oryx (*Oryx gazella*) in the Kalahari Desert, South Africa. (b) The oryx's carotid rete system cools the hot arterial blood from the body before it enters the brain.

Heat exchange for heating

Countercurrent heat exchange also occurs in animals living in extremely cold climates to reduce heat loss and maintain body temperature.

Penguins have heat exchangers in their flippers, feet and tails. These extremities have a relatively large surface area and are exposed to the cold, so they lose heat quickly. Blood from the feet flows back to the heart through veins close to the arteries. The warm blood in the arteries transfers heat to the veins so that blood moving back towards the heart is warmed, maintaining the penguin's body temperature (Figure 6.4.13). The blood travelling to the feet is cooled, so heat loss is minimised.

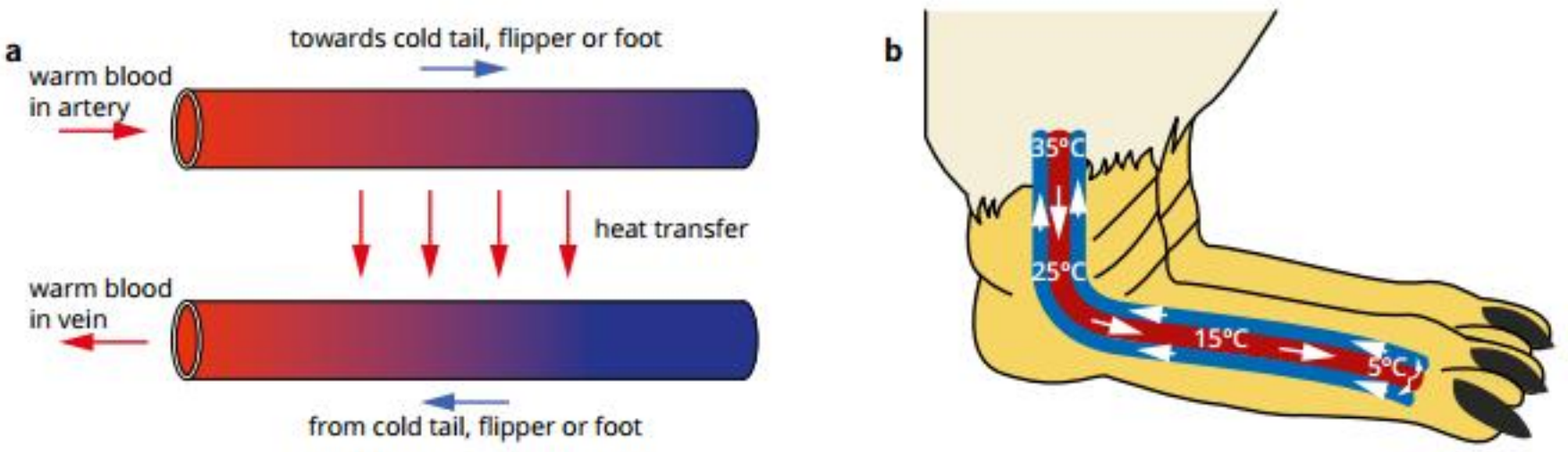


FIGURE 6.4.13 The heat exchange mechanism in the circulatory system of penguins ensures that heat loss at the extremities is minimised, while the core body temperature is maintained.

The diameter of the arteries flowing through the feet is also reduced to decrease the flow of blood to the extremities and further reduce heat loss. In this way, the cells in the feet receive oxygen and nutrients and remain warm enough to function, but less heat is lost to the environment.

THERMOREGULATION IN HUMANS

Humans have an array of physiological mechanisms to cope with extreme temperatures (Figure 6.4.14 on page 234), as do all endotherms. This section examines the human response to heat and cold.

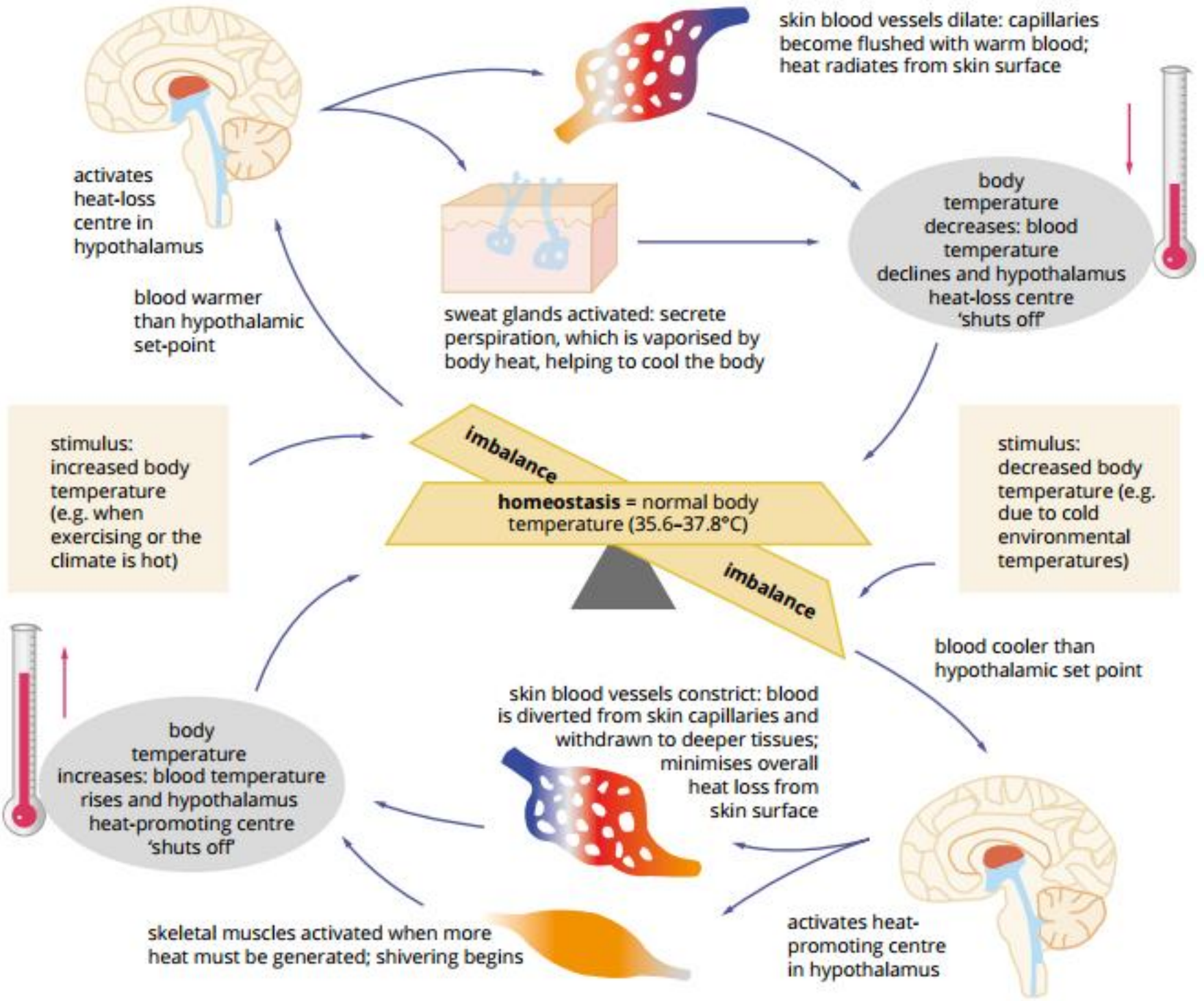

skin blood vessels dilate: capillaries become flushed with warm blood; heat radiates from skin surface
activates heat-loss centre in hypothalamus
body temperature decreases: blood temperature declines and hypothalamus heat-loss centre 'shuts off'
blood warmer than hypothalamic set-point
sweat glands activated: secrete perspiration, which is vaporised by body heat, helping to cool the body
stimulus: increased body temperature (e.g. when exercising or the climate is hot)
imbalance
homeostasis = normal body temperature (35.6–37.8°C)
imbalance
stimulus: decreased body temperature (e.g. due to cold environmental temperatures)
blood cooler than hypothalamic set point
skin blood vessels constrict: blood is diverted from skin capillaries and withdrawn to deeper tissues; minimises overall heat loss from skin surface
body temperature increases: blood temperature rises and hypothalamus heat-promoting centre 'shuts off'
skeletal muscles activated when more heat must be generated; shivering begins
activates heat-promoting centre in hypothalamus

FIGURE 6.4.14 Thermoregulation in humans involves a range of regulatory mechanisms that function to maintain thermal homeostasis (normal body temperature) for optimal functioning of the organism.

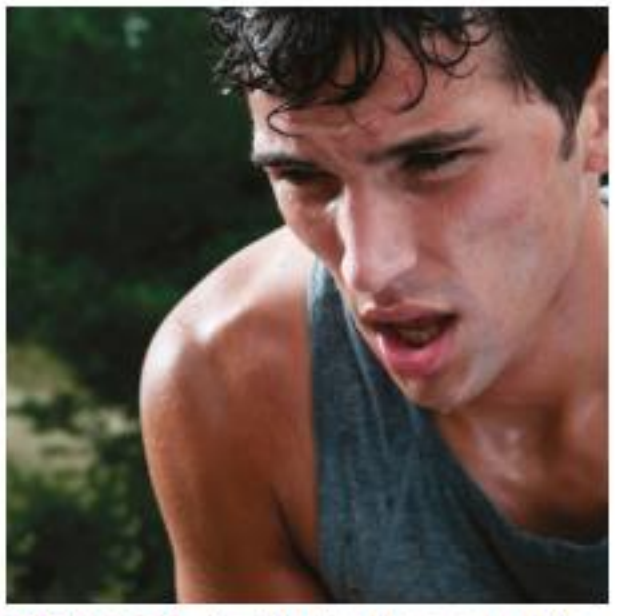


FIGURE 6.4.15 Sweating results in evaporative cooling and is one of the human body's homeostatic mechanisms to regulate body temperature.

Response to heat

Humans are one of the few animals that produce sweat to cool down. Evaporative heat loss (or evaporative cooling) is a very effective way of losing heat energy from the body. A change of state from liquid to gas is an endothermic process; that is, it requires an input of energy. In evaporative cooling of the skin, this energy comes from your body, in the form of heat energy (Figure 6.4.15).

Humans have two types of sweat glands: apocrine glands and eccrine glands. The function of apocrine glands is thought to be mainly scent or pheromone production, while the eccrine sweat glands function to control body temperature. These glands are distributed over most of the body and release sweat onto the skin surface through pores when your body temperature rises. These glands extend just below the surface of the dermis and secrete odourless sweat that is high in electrolytes and sodium. The rate of sweat secretion increases from almost zero in cold conditions to about 1.5 litres per hour in a hot environment. After about six weeks' acclimatisation to heat, this can increase to 4 litres per hour, which means a much greater evaporative cooling capacity is possible.

Some other voluntary and involuntary ways that your body responds to heat are:

- slowing the rate of cellular respiration in internal organs, which decreases heat generation, thus decreasing body temperature
- **vasodilation** (dilation of the blood vessels in the skin). Dilation means more blood is sent to the extremities. Heat is lost to the environment by radiation and convection (especially if it is windy)
- changes in behaviour such as:
  - removing clothing, which reduces the insulating effect of clothing layers and allows heat to escape from the skin
  - moving out of sunlight into shade
  - decreasing activity.

Figure 6.4.16 shows how an increase in temperature (internal or external) triggers negative feedback mechanisms to increase heat loss and decrease heat production.

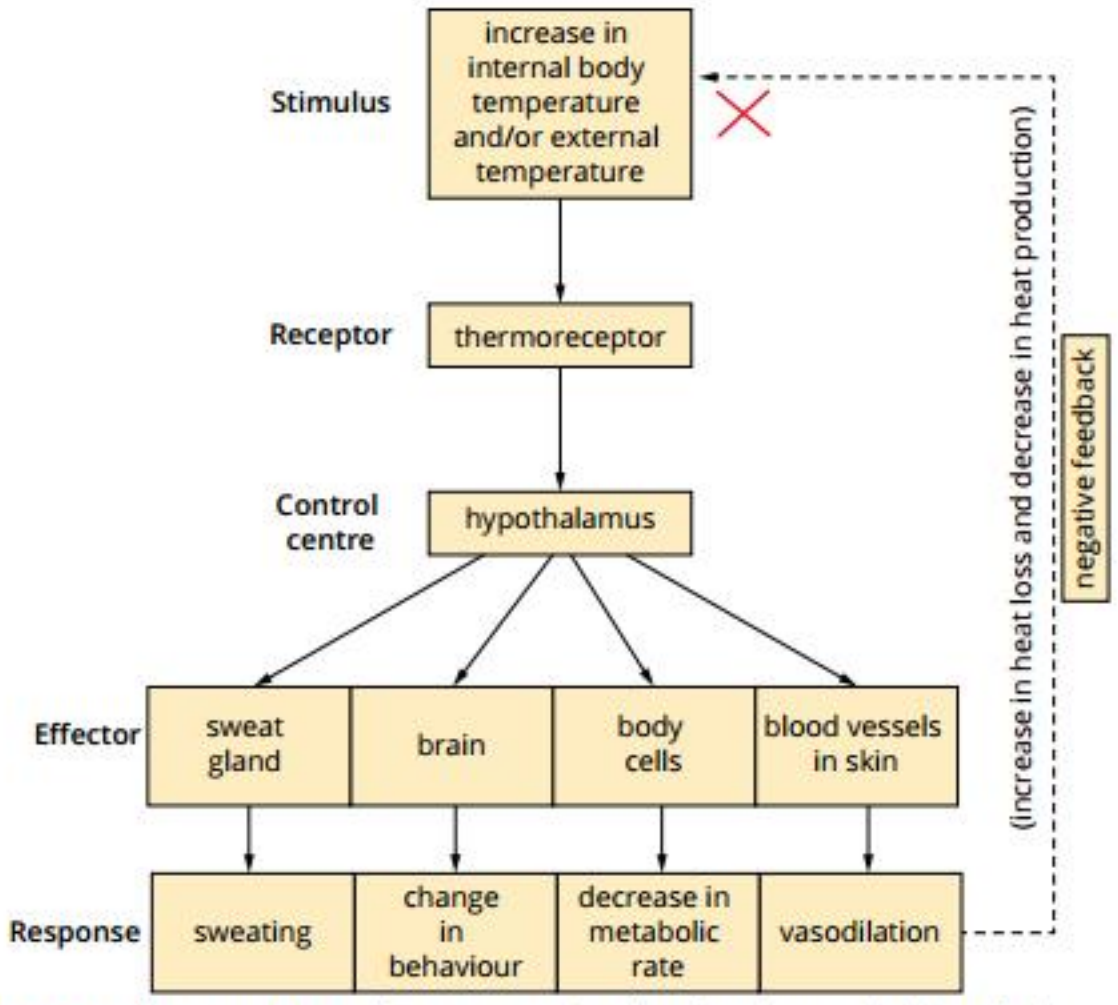


FIGURE 6.4.16 Human responses that can occur when there is an increase in internal body temperature and/or external temperature.

Response to cold

In humans, several nervous and endocrine responses occur rapidly to reduce heat loss from the body and to increase heat production when the body becomes too cold.

The following voluntary and involuntary responses reduce heat loss from the human body.

- **Vasoconstriction** (constriction of the blood vessels in the skin) reduces heat loss from the skin, as the amount of blood moving close to the exposed surface is reduced. This means that less body heat is lost to the environment.
- **Piloerection** is the constriction of the piloerector muscles around hair follicles ('goose bumps'), which increases the insulating effect of the hairs (Figure 6.4.17). This response has a minimal effect in humans but in animals with thick fur, the layer of trapped air increases significantly and reduces heat loss from the body.

FIGURE 6.4.17 A close-up of a human forearm with goose bumps. The contraction of blood vessels and small muscles (arrectores pilorum) that are attached to the base of each hair follicle pull the hair into an upright position. In this position, the skin resembles plucked goose skin.

- Changing body shape or decreasing surface area (e.g. curling up to make yourself small) reduces the area exposed to the cold and reduces the rate at which heat is lost from the skin.

The following responses increase heat production in the human body.

- Voluntary movement—during physical effort, the amount of heat produced by the muscles is increased.
- **Shivering thermogenesis**—the production of metabolic heat is increased through shivering. This involuntary movement of the muscles generates especially large amounts of heat. Shivering thermogenesis is stimulated by adrenaline.
- **Non-shivering thermogenesis** in brown fat—increased cellular activity in brown adipose tissue, which is a tissue specialised for heat production, causes the tissues to warm up. The heat produced is carried to other parts of the body in the blood.
- Increasing metabolism (the rate of cellular respiration)—metabolic processes in the internal organs are the main source of heat when the organism is at rest. In humans, around 60% of the energy released during cellular respiration is transformed into thermal energy. In humans, the overall metabolic rate, and therefore the rate of heat production, is controlled by hormones.

Figure 6.4.18 shows how a decrease in temperature (internal or external) triggers negative feedback mechanisms to decrease heat loss and increase heat production.

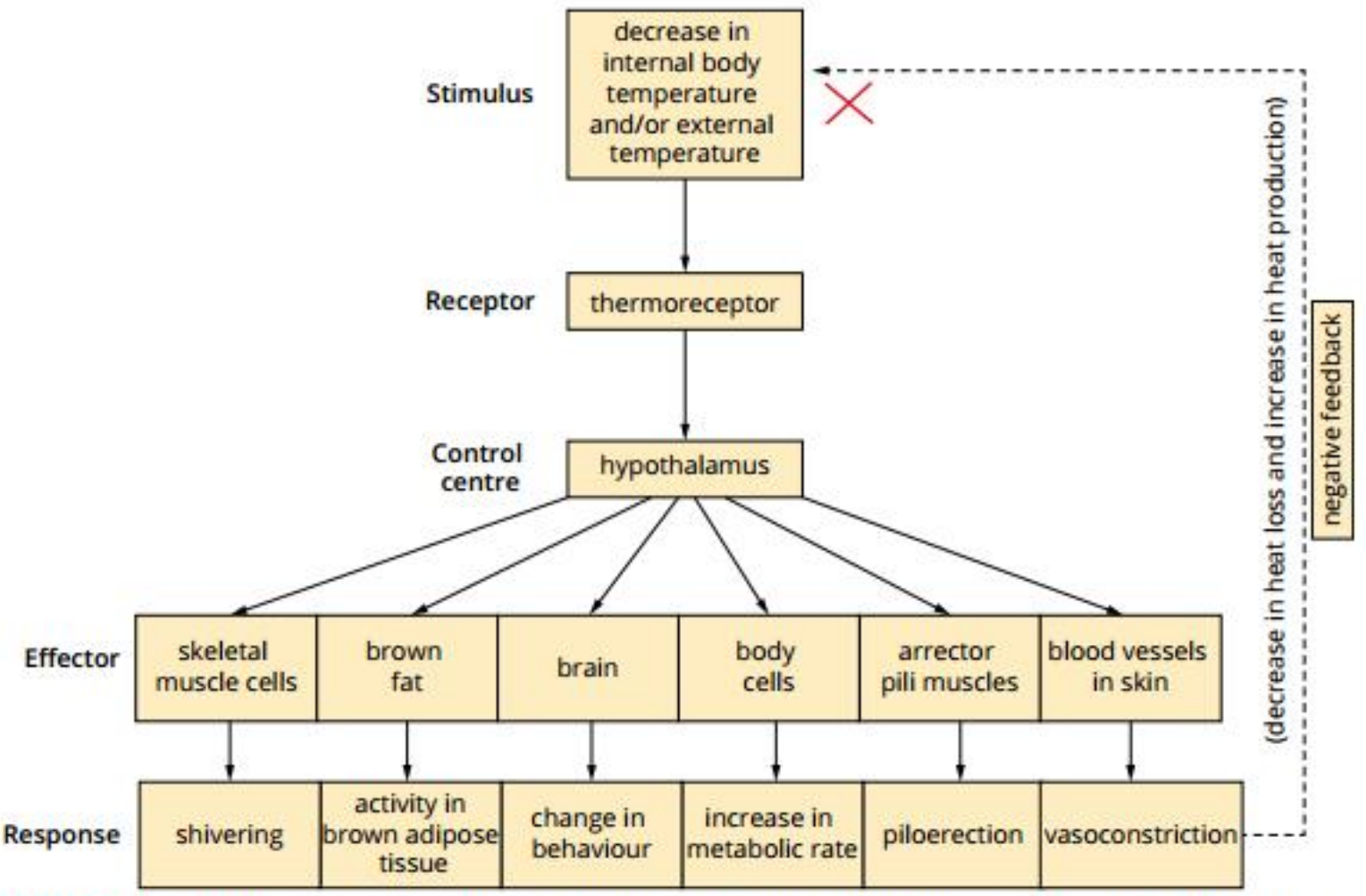


FIGURE 6.4.18 Human responses that can occur when there is a decrease in internal body temperature and/or external temperature

6.4 Review

SUMMARY

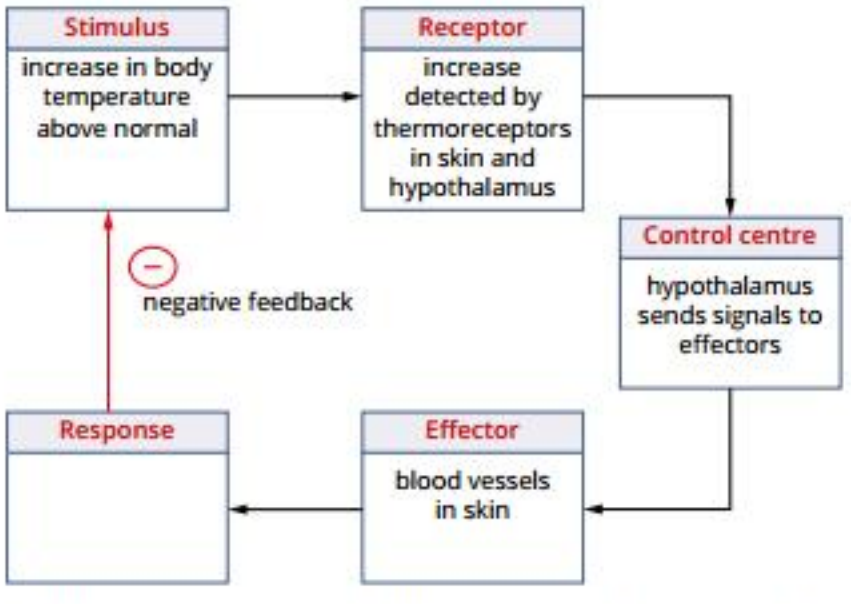
- Thermoregulation is when an organism regulates its body temperature either by increasing its metabolic activity or through behavioural responses.
- Endotherms are animals that can generate heat to maintain their body temperature using negative feedback loops.
- A change in the temperature of the hypothalamus initiates regulatory responses that can involve heat production or heat exchange.
- Temperature receptors (thermoreceptors) are found in the skin and hypothalamus.
- Heat is lost from the body by conduction, convection, radiation and evaporation.
- Structural adaptations of endotherms to conserve or release heat include:
  - body coverings and insulation, such as thick fur and blubber (fat)
  - large ears to increase heat loss and small ears to reduce heat loss
  - brown adipose tissue
  - increase mitochondria in cells.
- Behavioural responses of endotherms to conserve or release heat include:
  - kleptothermy (huddling for warmth)
  - torpor (hibernation and aestivation)
  - panting, licking skin, wallowing and gular fluttering
  - seeking shade or sunlight
  - nocturnal activity and burrowing.
- Physiological mechanisms of endotherms to conserve or release heat include:
  - counter-current heat exchange mechanisms
  - evaporative heat loss
  - thermogenesis (shivering thermogenesis and non-shivering thermogenesis)
  - vasomotor control (vasodilation and vasoconstriction).
- Stimulus–response models can be used to represent the mechanisms used to regulate temperature in different organisms.

KEY QUESTIONS

Describe

1 Recall three mechanisms humans use to produce heat.

2 Identify and fill in the missing response within the stimulus-response model below. The response is to an increase in core body temperature (use the effector as a clue).



3 Outline the role of brown adipose tissue in response to heat loss.

4 Identify and explain at least one structural feature, one physiological mechanism and one behavioural response that endotherms use to survive in cold temperatures.

5 All organisms exchange heat with their environment. Using examples, summarise and explain each of the four methods of heat exchange.

Apply

6 Draw a diagram showing a stimulus-response model that represents the homeostatic mechanisms that occur when there is a decrease in core body temperature and an increase in core body temperature.

7 An individual was hiking in the Blue Mountains on a very cold day in winter for an extended period of time. This person entered a state of hypothermia.

a What is meant by the term 'hypothermia'?

b Using a stimulus–response model, explain how one homeostatic mechanism acts to return the hiker's body temperature to normal.

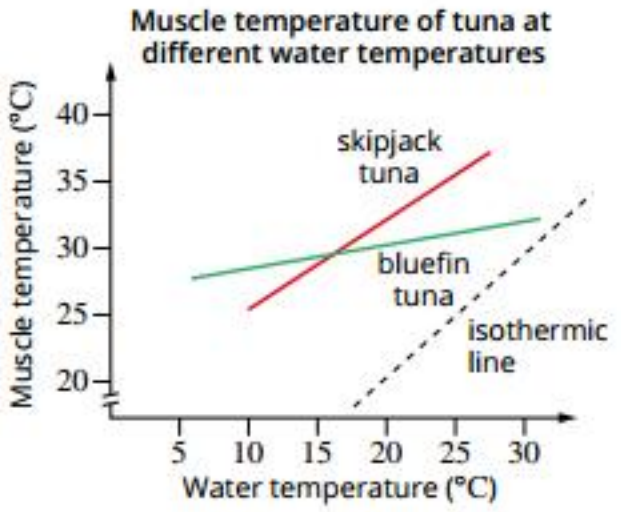
c Describe two physiological responses that allow for the regulation of body temperature to return to a normal range.

continued over page

6.4 Review *continued*

Analyse

8 Groups of two species of tuna (skipjack and bluefin) were kept in water at different temperatures. Maximum muscle temperature was measured in fish of each species taken from different water temperatures. The collected data is shown on the graph. The isothermic line represents tissue temperature equivalent to water temperature. If the muscle temperature of a fish is parallel to the isothermic line, it means that the muscle temperature is a constant value above the ambient water temperature.



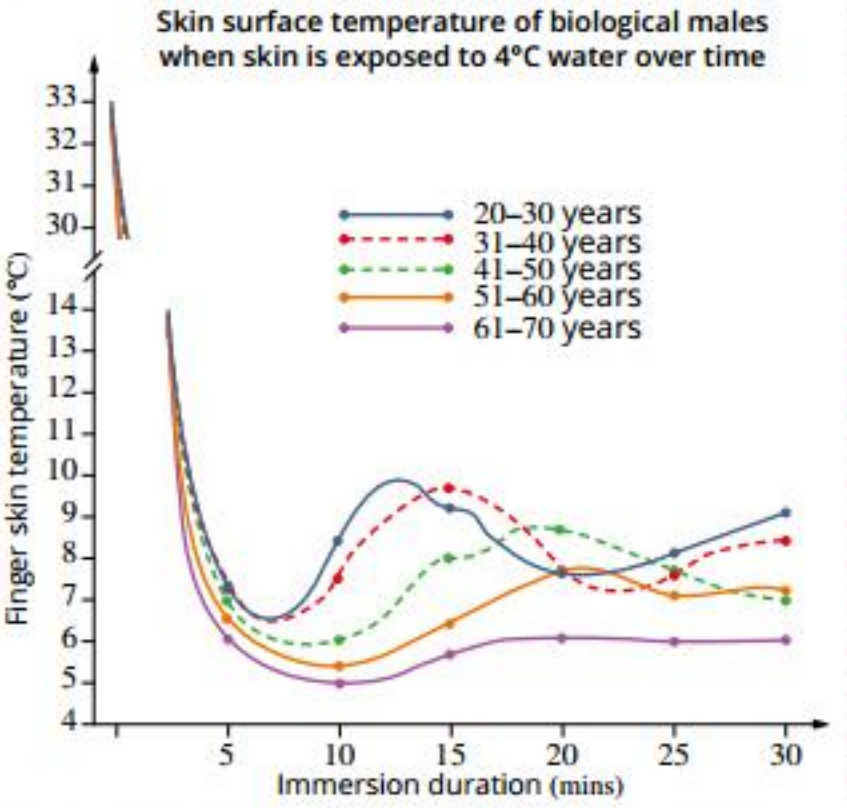
a Identify the muscle temperature for the following fish at a water temperature of 25°C:
- **i** skipjack tuna
- **ii** bluefin tuna.

b **i** Explain the difference between an endotherm and an ectotherm.
- **ii** Fish are ectotherms. Infer why there is such a difference between muscle temperature and water temperature for each species.

9 Occasionally an individual is born without sweat glands and cannot lose heat or water through their skin. A person without sweat glands and a person with normal sweat glands were placed in cool, dry conditions and their skin and mouth temperatures were recorded. The two people were then placed in a moist, hot environment, and further recordings were made. The results of the experiment are recorded in the table above. Deduce which person (A or B) was born without sweat glands. Explain your reasoning.

Response	Person A		Person B	
	Cool, dry	Hot, moist	Cool, dry	Hot, moist
skin temperature (°C)	33.8	40.5	32.7	37.4
oral temperature (°C)	36.9	38.6	36.8	37.2
water loss from skin and lungs (mL)	not recorded	20.0	not recorded	282.0
urine volume (mL)	not recorded	280.5	not recorded	12.6

10 The graph below shows the variation in finger skin surface temperature when the finger of biological males at different ages is immersed in ice water (4°C) over a period of time.



a Describe what type of receptors detect change in temperature.

b Explain the skin temperature changes in terms of vasodilation and vasoconstriction.

c With extended exposure to ice water, the skin surface temperature settles at a temperature well below the starting core body temperature. State the temperature that the skin appears to settle at for the 61–70 year old group.

d Compare the final skin temperature between the younger group tested (20–30 years) and the oldest group tested (61–70 years). Suggest a reason as to why this variation might exist.

6.5 Osmoregulation

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain osmoregulation in humans, including the role of antidiuretic hormone (ADH) and the kidneys using feedback control diagrams
- explain how structural and homeostatic mechanisms maintain water balance in plants, including the roles of stomata, vacuoles, cuticle and abscisic acid
- interpret data from an experiment comparing the number and distribution of stomata in plants adapted to different environments.

OSMOREGULATION IN HUMANS

Humans rely on their external environments to provide adequate levels of nutrients, water and oxygen and suitable physical conditions, such as light and temperature. Water plays a key role in the human body and it is regulated via the process of osmoregulation. Humans have a range of mechanisms that allow them to adapt to changes in water balance while maintaining a stable internal environment when external conditions fluctuate.

Maintaining water balance is necessary to control salt concentrations. Salts form ions in solution, and cells require the concentrations of ions to be held within narrow limits for biochemical processes to occur efficiently. Moreover, if internal salt concentrations are too high, water will move by osmosis out of cells into extracellular fluid (Figure 6.5.1). Some ions (such as the hydrogencarbonate ion) are also important for regulating the pH of body fluids, which must be at a suitable pH for enzymes and other molecules to function efficiently. Maintaining the correct concentrations of ions is achieved by regulating both water and salt balance.

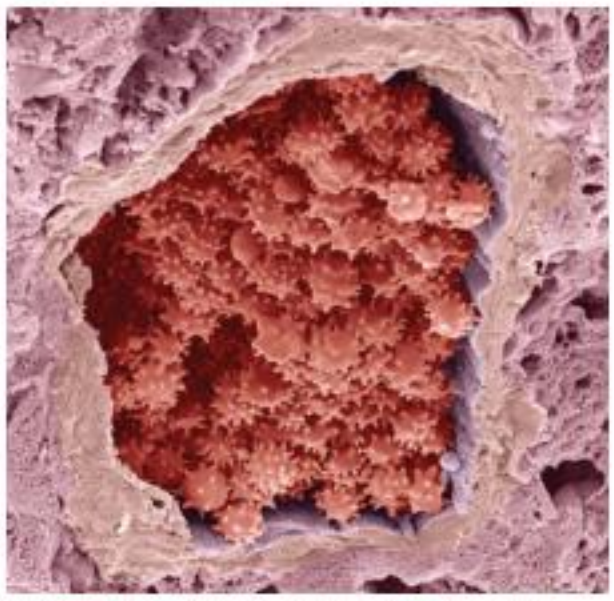


FIGURE 6.5.1 The maintenance of constant osmotic pressure in the blood is important because it prevents red cells from dehydrating or bursting. This scanning electron micrograph of a section through an arteriole shows crenated (wrinkled) red blood cells caused by dehydration, which distorts the red blood cell.

Water balance involves regulating the intake and loss of both water and salts. In organisms, net movement of water into and out of cells occurs as a result of osmosis, which is regulated by solute concentrations. Water moves across a semipermeable membrane from regions of lower solute concentration (higher concentration of free water molecules) to regions of higher solute concentration (lower concentration of free water molecules).

The amount of water lost or gained throughout the day differs between individuals and depends on the amount of exercise, temperature, humidity, food and fluid intake. Urination rather than water intake is a better indicator of whether an individual has good water balance. A healthy person with adequate hydration usually urinates 4–8 times per day, and the urine is pale yellow.

Water gain and loss

The total volume of fluid taken into the body depends on diet and activity levels, and typically varies from about 2 to 16 litres per day. The minimum water requirement for fluid replacement in a 70 kg person in a cool climate is about 3000 mL per day. Of this, about 400–600 mL is obtained by eating, and about 400 mL is produced by aerobic respiration. (This is called metabolic water because it is produced in cellular respiration.) The remainder of about 2000–2200 mL must be obtained by drinking.

For a 70 kg person, water will be lost mainly in urine (500–1500 mL per day), evaporation from the respiratory system (400–800 mL per day), sweat (100–800 mL per day), and faeces (100–200 mL per day).

Hormonal control of water balance

Water and solute concentration are monitored by **osmoreceptors** in the hypothalamus and **baroreceptors** in the atria of the heart. Osmoreceptors are sensitive to blood solute concentrations, while baroreceptors detect changes in blood pressure, which is an indication of the volume of blood. Collectively, these receptors detect the solute concentration in blood and extracellular fluid. The unit of measurement used for these blood solute concentrations is osmolality, because they contribute to osmotic effects on cells.

Because cell membranes are permeable to water, the **osmolality** in the extracellular fluid is approximately the same as it is in the intracellular fluid (cytosol). Changes in the osmolality of the extracellular fluid will therefore affect cytosol concentrations, which can cause problems with cellular metabolic reactions. Compared to extracellular fluid, the cytosol of cells is high in potassium and magnesium and low in sodium and chloride ions.

Osmolality is a measure of the concentration of particles (such as sodium ions and chloride ions) that affect osmosis.

Antidiuretic hormone (ADH), also called vasopressin, regulates water reabsorption. It is synthesised in the hypothalamus and transported to the posterior pituitary gland, where it is stored (Figure 6.5.2). When osmoreceptors in the hypothalamus detect an increase in the osmolality of the blood, a signal is sent to the posterior pituitary gland, and ADH is released.

ADH acts on the kidneys to increase the permeability to water of the distal tubules and collecting ducts. The collecting ducts run through the medulla of the kidney, which has high salt levels (and therefore a higher osmotic potential). This causes the absorption of water from the tubules back into the blood by osmosis, decreasing urine output; the urine becomes more concentrated and has a darker yellow colour. As the blood returns to a normal concentration, negative feedback stops the production of ADH.

Conversely, if the osmoreceptors detect a decrease in osmolality (e.g. if too much water has been taken into the body), ADH release will be decreased. This reduces the reabsorption of water and consequently increases urine volume; the urine becomes more dilute and has a paler yellow colour (Figure 6.5.2).

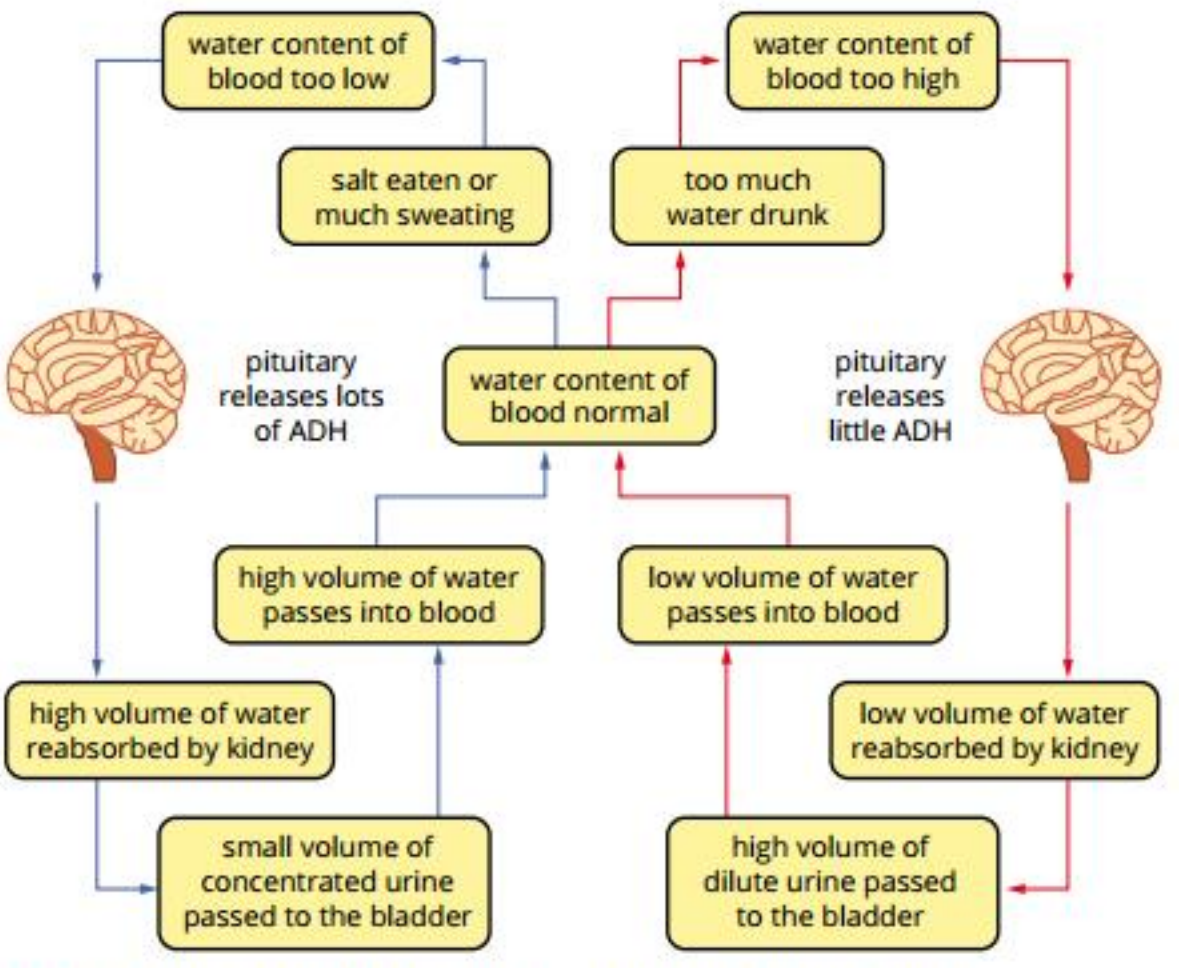

water content of blood too low
water content of blood too high
salt eaten or much sweating
too much water drunk
pituitary releases lots of ADH
water content of blood normal
pituitary releases little ADH
high volume of water passes into blood
low volume of water passes into blood
high volume of water reabsorbed by kidney
low volume of water reabsorbed by kidney
small volume of concentrated urine passed to the bladder
high volume of dilute urine passed to the bladder

FIGURE 6.5.2 Hormonal control of water balance by antidiuretic hormone (ADH)

A number of substances such as nicotine, alcohol and narcotics can interrupt the feedback control of water balance in the body. This can also occur because of pain, stress or hypothermia (lowered body temperature).

Changes in blood osmolality or blood pressure also stimulate counteracting response. Initially an enzyme called **renin** is secreted from the kidneys in response to these changes (Figure 6.5.3). Renin then triggers a series of reactions involving other hormones that results in the release of **aldosterone** from the adrenal glands situated above the kidney. Aldosterone simultaneously regulates sodium and potassium levels by increasing potassium excretion into the urine and causing sodium reabsorption into the blood. This causes more water to be drawn into the blood by osmosis, thus increasing blood volume and pressure.

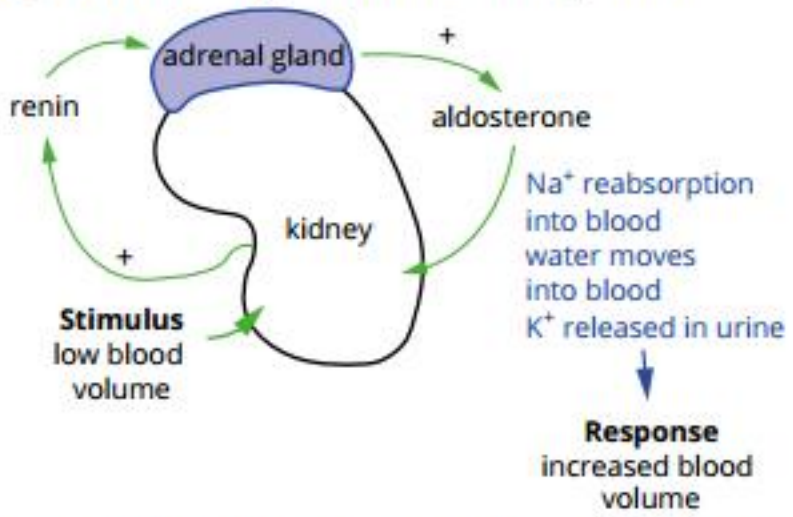


FIGURE 6.5.3 Hormonal control of blood volume. The stimulus for this negative feedback loop is low blood volume.

A lack of aldosterone can result in low sodium levels, high potassium levels and high acid levels in the blood. These are potentially dangerous conditions. People with an aldosterone deficiency suffer from Addison's disease and must take a synthetic hormone called fludrocortisone acetate to manage this condition.

Osmoregulation in plants, like humans, is needed to regulate water balance throughout the organism. This process is essential to enable survival in changing environments. Water loss in plants occurs through the process of **transpiration**, in which water is lost from the plant's leaves by osmosis through small openings called **stomata**. The stomata help to regulate water loss while still permitting the entry of carbon dioxide gas needed by the plant for photosynthesis. Osmoregulation is an important aspect of maintaining homeostasis in plants as shown in Figure 6.5.4. If an organism is not able to adapt to its external environment, it will suffer cellular damage and possibly death when conditions change.

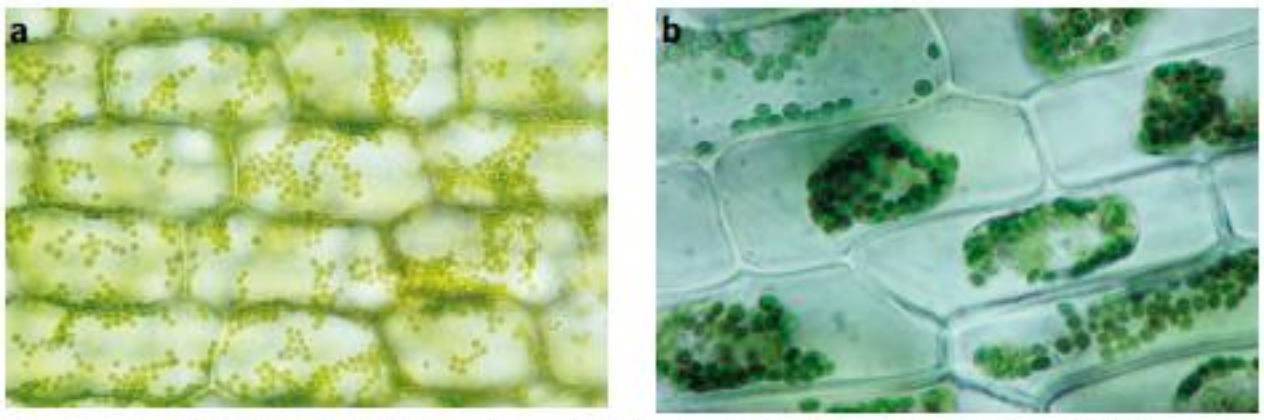


FIGURE 6.5.4 The internal cellular environment of the freshwater Canadian pondweed (*Elodea* sp.) responds to changes in the external environment using osmosis. (a) In fresh water, chloroplasts move freely through the cytoplasm, as solute concentration is equal in both the internal and external environments. (b) When the cells are exposed to a salty environment, water leaves the cell by osmosis and the cell membrane contracts, clumping the chloroplasts in the middle of the cell. Because *Elodea* is a freshwater plant, it does not have the regulatory mechanisms needed to survive in a saltwater environment.

Plant hormones

Plant hormones, like all signalling molecules, are produced in low concentrations but have a significant effect on plant development and growth. In contrast to animal hormones, which are produced by specific glands and organs, each plant cell can produce many different types of hormones. Plant hormones also have a variety of modes of transmission.

There are five main types of plant hormones: **abscisic acid (ABA)**, **auxin**, **cytokinins**, **ethylene** and **gibberellins**. These are sometimes called **phytohormones** (from the Greek *phyton*, meaning 'plant'). A summary of the biological roles of these hormones is provided in Table 6.5.1.

TABLE 6.5.1 The biological roles of the five main types of plant hormones

Type of plant hormone	Source	Effector site	Mode of transmission	Visible effect	
Abscisic acid (ABA)	leaves (chloroplasts), roots	seeds, buds, guard cells, leaves and fruit	transported in the xylem from roots and phloem from leaves	seed and bud dormancy, drought tolerance and apical dominance, induces stomatal closure	
Auxin	shoot tip (meristem), seeds	growing region of shoots and roots, developing fruit	transported from cell to cell, often with directional transport, usually moving from shoots towards roots	shoot tip bends towards the light (phototropism), roots grow downwards (gravitropism), apical dominance	
Cytokinins	roots and developing fruits	branch and leaf buds	transported in xylem	growth of lateral branches	
Ethylene	ripening fruits and other parts of the plant	most cells	diffusion (ethylene is a gas)	increases sugar content of fruit, fruit and leaf drop	
Gibberellins	root and shoot apical meristems, growing leaves and seeds	meristems, leaves, seeds and flowers	typically used in the cell that made it, otherwise transported cell-to-cell in xylem and phloem	elongation of stems, leaf expansion, seed germination, fruit and flower maturation	

Structural features of plants

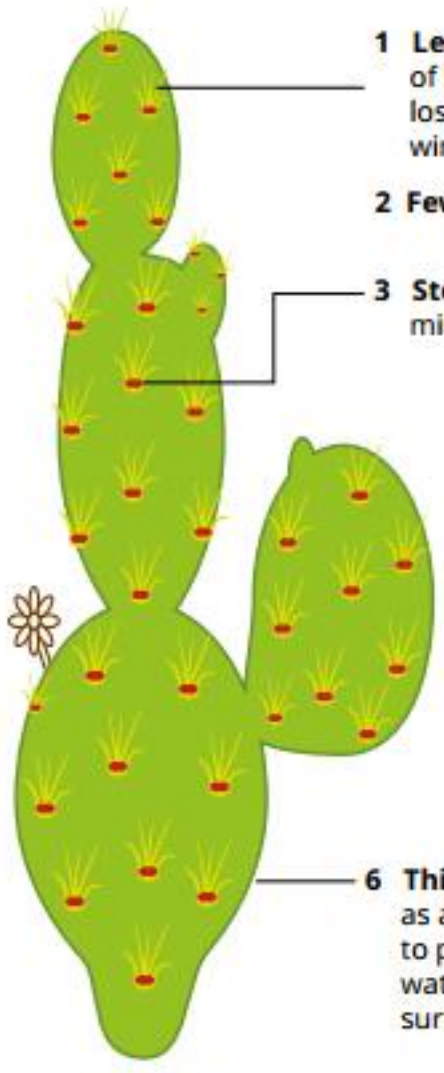
Water is essential for photosynthesis. Therefore, a large number of structural features that we observe in plants reduce water loss caused by salinity, heat and wind in the environment. These features include:

- leaf surface area
- few stomata
- stomatal hairs that create a humid microclimate
- sunken or protected stomata
- thick, waxy cuticle
- extensive root systems
- rolled leaves
- leaves orientated away from the sun.

Plants that grow in dry, hot environments are known as **xerophytes** (from Greek *xeros*, meaning 'dry', and *phyton*, meaning 'a plant'). Some of their adaptations are discussed in more detail below.

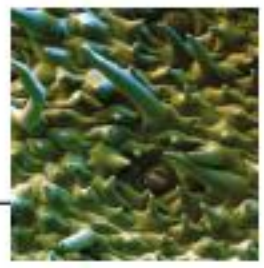
Cacti are well-known xerophytes. Xerophytes have structural features that conserve moisture and prevent the leaf temperature from rising too much. They also have an increased tolerance to desiccation (drying). Some of the adaptations of xerophytes are shown in Figures 6.5.5 and 6.5.6.

FIGURE 6.5.5 A scanning electron micrograph of the sunken stomata of a needle leaf of the coastal sitka spruce (*Picea sitchensis*).



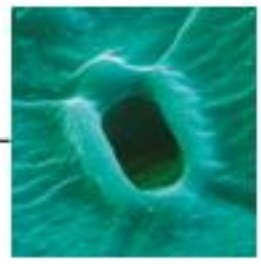
Prickly pear

1 **Leaf spines** (modified leaves) reduce the surface area of leaves and number of stomata, helping to slow the loss of water from the plant in response to hot and windy environments.



2 **Few stomata** (pores) in leaves reduces transpiration.

3 **Stomatal hairs** covering leaves create a humid microclimate that helps reduce transpiration.



4 **Sunken or protected stomata** create a humid microclimate around the stomata and minimise water loss.

5 **Rolled leaves** maintain humid air around the stomata.

6 **Thick, waxy cuticles** act as a water retention layer to prevent the diffusion of water molecules from the leaf surface in response to heat.

7 **Extensive roots** maximise water uptake.

FIGURE 6.5.6 Plants that live in harsh, dry environments such as deserts have evolved features that enable them to conserve water.

Marram grasses (*Ammophila* species) are xerophytes that grow well in the salty, sandy soils of coastlines (Figure 6.5.7a). When conditions are hot and dry, thin-walled bulliform (bubble-shaped) cells partially collapse, causing the leaves to roll inwards, reducing water loss. Hairs on the inside of the rolled-up leaf trap moisture, creating a humid microclimate (Figure 6.5.7b). This humidity reduces the concentration gradient between the outside and inside of the leaf, which reduces transpiration. Because of this and other adaptations, these grasses can survive in and then stabilise sand dunes that are prone to erosion.

FIGURE 6.5.7 (a) Marram grasses (*Ammophila* species) grow well in the salty, sandy soils of coastlines. They have features and mechanisms that allow them to survive in this dry, salty environment. (b) One of these mechanisms is leaf rolling. This enables the plant to trap moisture and reduce water loss.

Homeostatic mechanisms in plants

As mentioned above and listed in Table 6.5.1 (page 242), there are five major groups of plant hormones. Plant hormones are involved in the loss of plant organs, the process known as abscission. The three hormones involved in abscission are ethylene, auxin and abscisic acid (ABA). When production of these hormones is stimulated by a lack of water, saline soils, cold temperatures or frost, it signals to plant organs that they will be undergoing some sort of stress, and then stimulates responses that help the plant protect itself. This is a stimulus-response pathway in plants.

With a focus on osmoregulation, the plant hormone, ABA, can be used as an example in regulating water balance. When plants experience water stress (a lack of water), a stimulus-response occurs that closes the leaf stomata and as a result, water loss by transpiration is prevented. ABA is the hormone synthesised in this process and triggers a series of responses that regulates the closing of stomatal pores by becoming increasingly flaccid.

When the stomata are open, the guard cells are turgid, or swollen. This turgidity is caused by the accumulation of K^+ (potassium ions) in the guard cells. As K^+ levels increase in the guard cells, the water potential of the guard cells drops, and water enters the guard cells. The release of ABA results in the loss of K^+ from guard cells and the stomata become flaccid. This inhibits the opening of stomata, so when ABA is present in guard cells the stomatal pores will remain closed. (Figure 6.5.8).

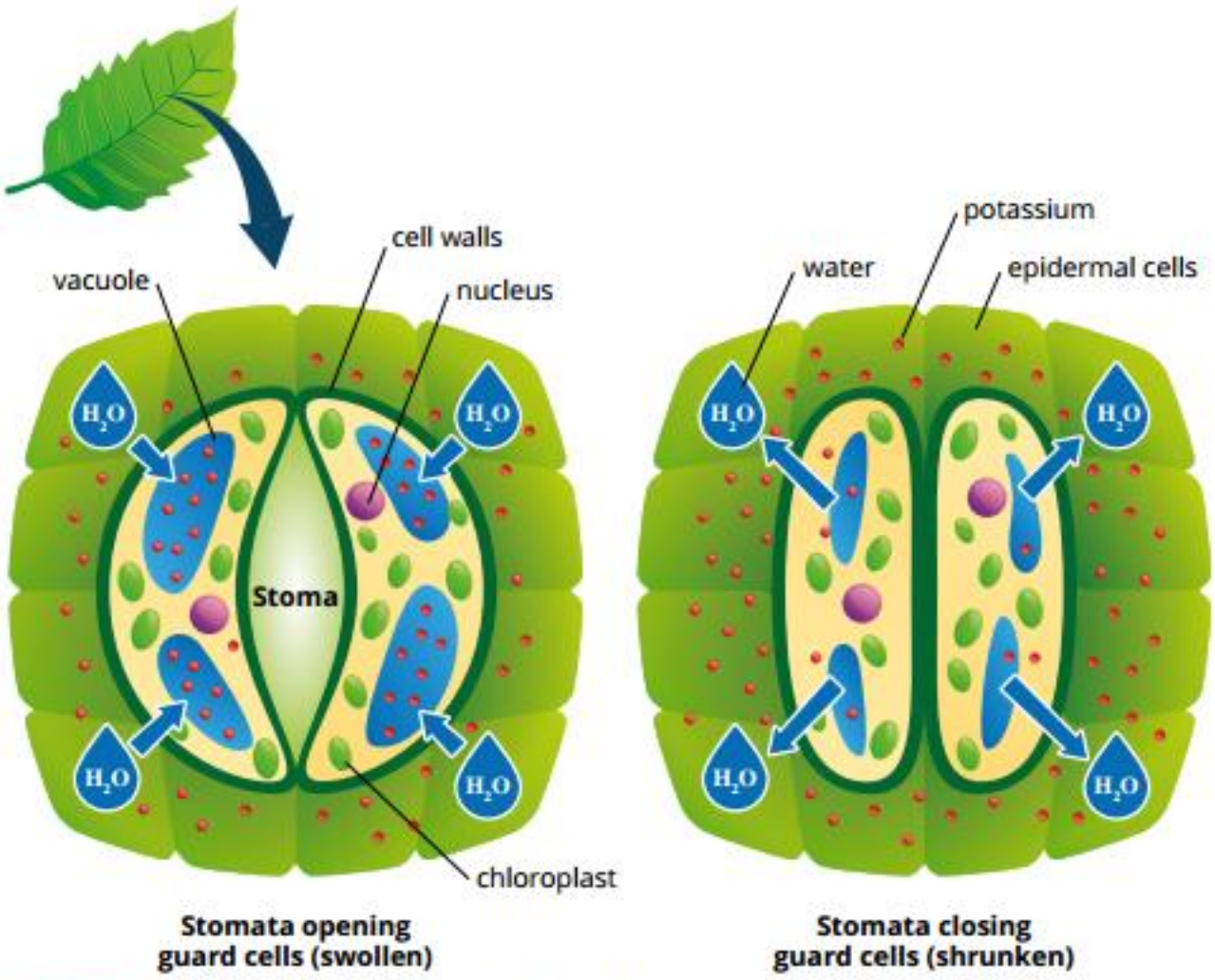


FIGURE 6.5.8 Stomata play an important role in regulating water loss from plants. When acted on by ABA the guard cells become flaccid, making the stomata close and reducing water loss via transpiration.

The action of ABA to induce stomatal closure during low water levels within a plant can be represented using a stimulus–response model. A stimulus–response model can be used to describe how plants restore their internal environment when there are fluctuations in their external environment. An example of a stimulus–response model in plants to low water levels is shown below:

- stimulus—reduced water level in plant.
- receptor—osmoreceptors in the mesophyll cells detect low water levels.
- effector—release of the plant hormone ABA, which triggers the release of potassium from guard cells.
- response—decreasing water pressure within the cells (reduced turgor). A loss of turgor makes the stomatal pore close, as the guard cells become flaccid and block the opening.

Using another example, in some plants, receptors in the roots respond to the drying out of the soil by releasing ABA into the xylem tissue. The secreted ABA then moves with the root-absorbed water to the leaf epidermis. Under continued drought conditions, ABA can also cause the dropping off, or abscission, of flowers, immature fruits and leaves. It does so by causing the disintegration of a special layer of cells (abscission zone) that are located at the base of the plant structures. Shedding of leaves reduces the surface area exposed to dry air and slows the water loss in the plant (Figure 6.5.9).

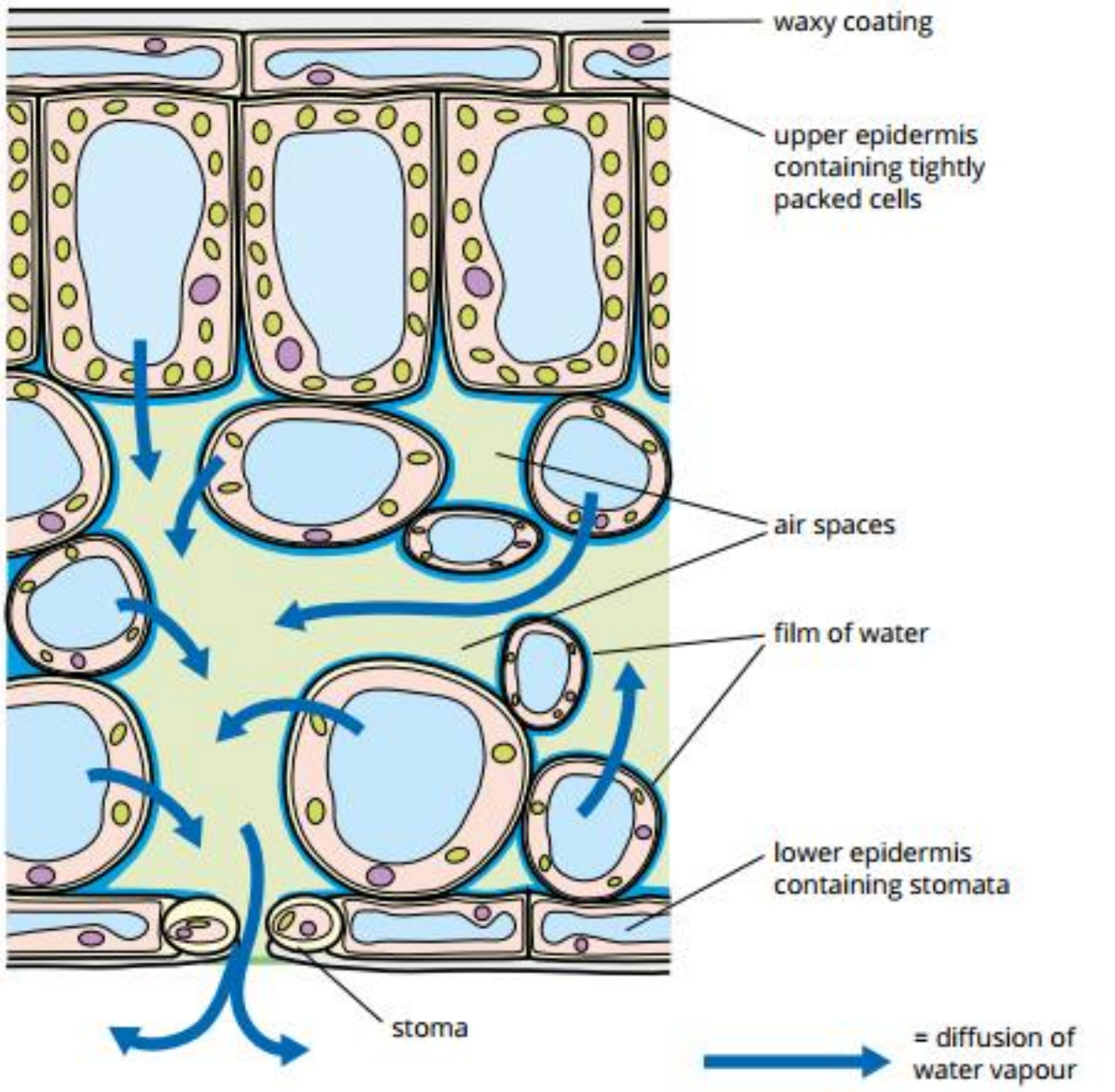


FIGURE 6.5.9 Under drought conditions, abscisic acid released from the plant's roots into the xylem makes its way up to the leaf epidermis and causes the closure of the leaf stoma. This prevents the loss of water vapour by simple diffusion.

Physiological mechanisms in plants

Plants inhabit an incredible range of environments, from the hottest deserts to high mountain peaks, fast-flowing rivers and even the coastal intertidal zone. Thus, they need an equally impressive range of physiological mechanisms to cope in what are often stressful conditions. These mechanisms play an important role in enabling plants to meet environmental challenges.

Crassulacean acid metabolism, also known as CAM photosynthesis (see Module 3.4), is an example of a physiological adaptation that enables improved efficiency in water storage and use in plants. It is most commonly found in plants living in dry environments, such as succulent plants in deserts. Some xerophytes and some plants adapted to saline conditions can minimise water loss during the heat of the day by using the CAM metabolic pathway.

In CAM plants, the stomata open only at night to collect carbon dioxide. Rather than using the carbon dioxide immediately, as non-CAM photosynthesising plants do, it is stored as malic acid in cell vacuoles. During the day, the malic acid is transported to the chloroplasts, where it is used to produce the carbon dioxide needed for photosynthesis (Figure 6.5.10). By storing the carbon dioxide required for photosynthesis at night, the plant is able to close its stomata during the heat of the day to reduce water loss. This physiological mechanism allows plants to survive in environments of extreme heat and aridity.

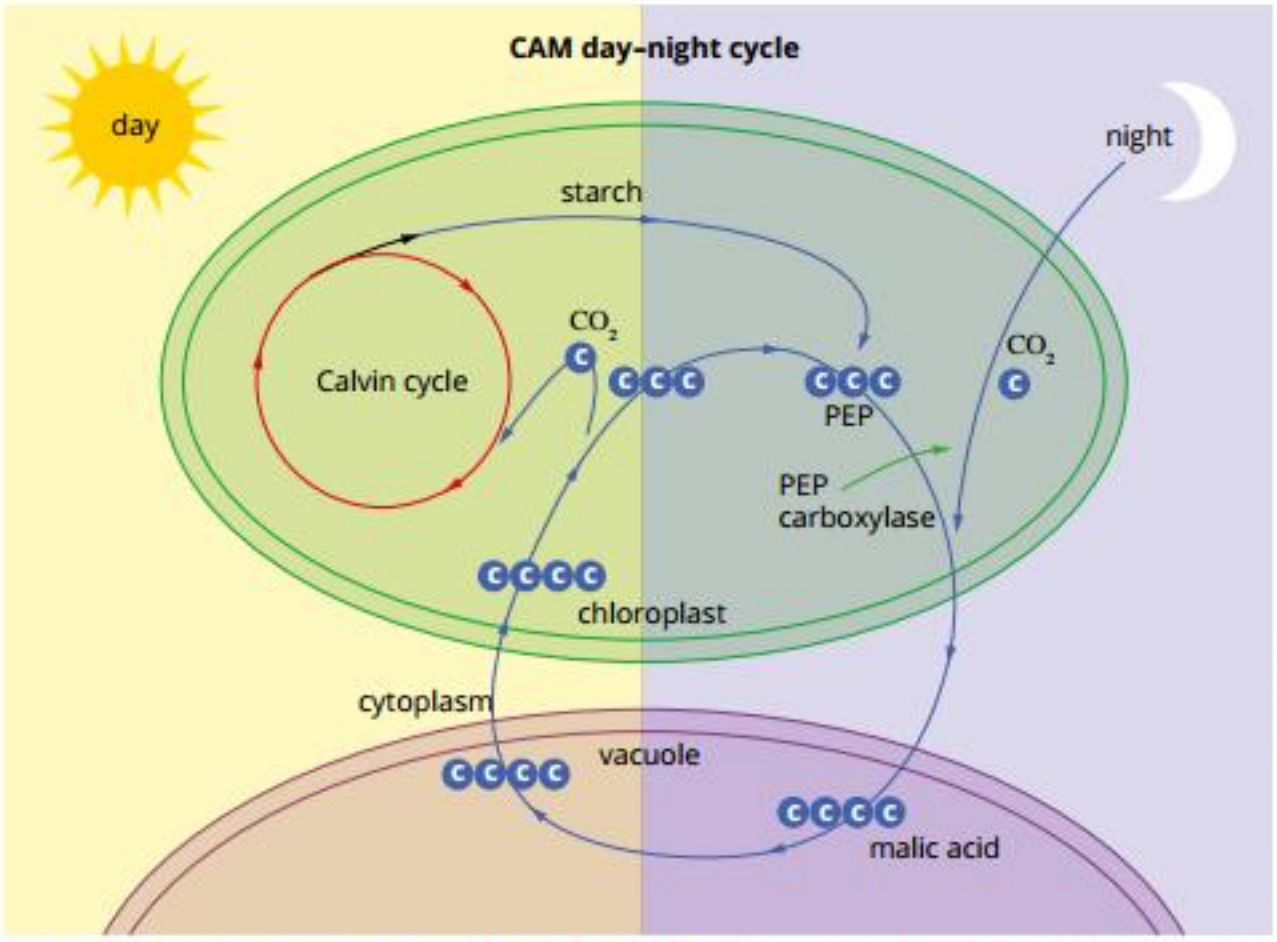


FIGURE 6.5.10 A summary of the complex CAM metabolic pathway of some plants in hot and dry or saline environments. This metabolic pathway enables the plants to absorb and store carbon dioxide at night to avoid losing precious water during the day. 'C' denotes the number of carbon atoms in each molecule.

Hydrophytes

So far, the emphasis has been on plants subjected to severe conditions involving a lack of water, high temperature or high salinity. Another group of plants, called **mesophytes**, have features that allow them to thrive in moist habitats and require well-aerated soils. These plants have fibrous roots and the ability to produce rhizomes, corms or bulbs for water and food storage.

But there are a group of specialised plants known as **hydrophytes** that only live in water, and their need is for CO_2 and light for photosynthesis. These plants, which include water hyacinths and *Elodea*, have generally got thin or reduced cuticles and a reduced root system. Stomata in land-based plants are mainly on the underside of the leaves, but in hydrophytes they are present only on the upper surface of the floating leaves. Submerged leaves generally do not have many stomata. These plants will also have air sacs and less vascular tissue. This assists their flotation to the surface, or as close to it as possible, in order to be exposed to as much sunlight as possible (Figure 6.5.11).

FIGURE 6.5.11 Hydrophytes are plants specially adapted to living in or under water.

6.5 Review

SUMMARY

- Water enters body cells throughout the day from: drinking, eating and cellular respiration.
- Salt is gained from our diet.
- Water is lost from the body mainly as urine, in faeces, across the skin and from the lungs.
- Salt is lost through the skin, kidneys and faeces.
- Osmoreceptors in the hypothalamus and baroreceptors in the atria of the heart detect the osmolality of the blood.
- An increase in osmolality causes a release of ADH from the pituitary, which acts on the kidney to increase water absorption back into the blood.
- As a result, osmolality of the blood decreases and the blood volume increases, and urine concentration increases and urine volume decreases.
- A decrease in osmolality causes decrease in ADH levels and urine volume will increase.
- Low blood volume stimulates the secretion of aldosterone.
  - Renin is secreted from the kidneys.
  - Renin causes release of aldosterone.
  - Aldosterone causes absorption of sodium into the blood.
  - Aldosterone causes potassium excretion into the urine.
- As a result of this multiple hormone action blood volume and blood pressure increase.
- Structural adaptations of plants include
  - reduced leaf surface area
  - fewer stomata
  - stomatal hairs to create a humid microclimate
  - sunken or protected stomata
  - thick, waxy cuticle
  - extensive root systems
  - leaf shape: rolled leaves; reduced surface area
  - leaves orientated away from sunlight.
- Physiological adaptations of plants include CAM, salinity tolerance (halophytes), drought tolerance (xerophytes) and water abundance (hydrophytes)
- Plants also have sensory receptors, which include photosensitive pigments (light intensity and wavelength detection), gravity and pressure sensors, and the ability to respond to touch by creating action potentials along cell membranes.
- Plants respond to dry soil conditions by their root cells secreting abscisic acid (ABA), which stimulates the closing of stomata.
- Plant hormones
  - are produced in a variety of cells in plants
  - can be hydrophilic or hydrophobic
  - are transported cell-to-cell or via the xylem and/or phloem.

KEY QUESTIONS

Describe

1 Describe three ways in which humans are able to gain water.

2 **a** Describe what osmolality measures.
b Identify two receptors that detect changes in osmolality in the blood.

3 Identify the part of the body that produces ADH and the part of the body that releases ADH.

4 Identify and list structural features of xerophytic plants.

5 What is the role of guard cells and stomata in regulating water loss in plants?

Apply

6 Draw or list the parts of a stimulus-response model which demonstrate a decrease in water balance in the body. Include the stimulus, receptor, control centre, effector and response.

7 Using a well-annotated diagram, represent the process of hormonal control of sodium and potassium levels by renin and aldosterone.

Analyse

8 An investigation was conducted to analyse the number of stomata present in different populations of plant species, each of which lives in a different environment. The number of stomata on the upper surface of a leaf was counted for each different plant species, determined by counting the stomata visible under a microscope. This was then repeated for the lower surface of the leaf and all results were presented in the table below.

	Number of stomata		Number of stomata		
Type of Leaf	**Upper leaf**	**Stomatal index (SI)**	**Lower leaf**	**Stomatal index (SI)**	**Upper : lower SI ratio**
Spinacia oleracea	7	4.76	13	11.50	0.41
Rhododendron spp.	0	0	9	6.04	0
Vinca minor	0	0	13	9.09	0
Epipremnum aureum	0	0	8	11.76	0
Hedera spp.	0	0	26	7.98	0

a Which plant has the highest stomatal index (SI) for the lower leaf?

b Which plant has the highest Upper : Lower SI ratio?

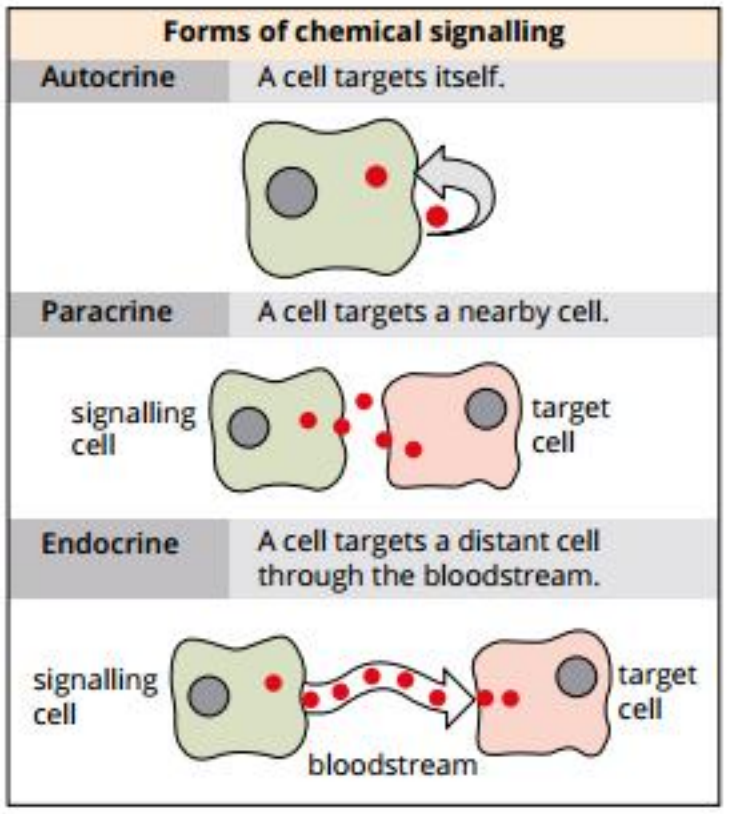
c Where are the majority of stomata located in a leaf from these species? And suggest why this might be the case.

d Outline the benefits of the CAM pathway in plants. Describe the environment CAM plants are adapted to, how they regulate their stomatal opening and their need to regulate water loss.

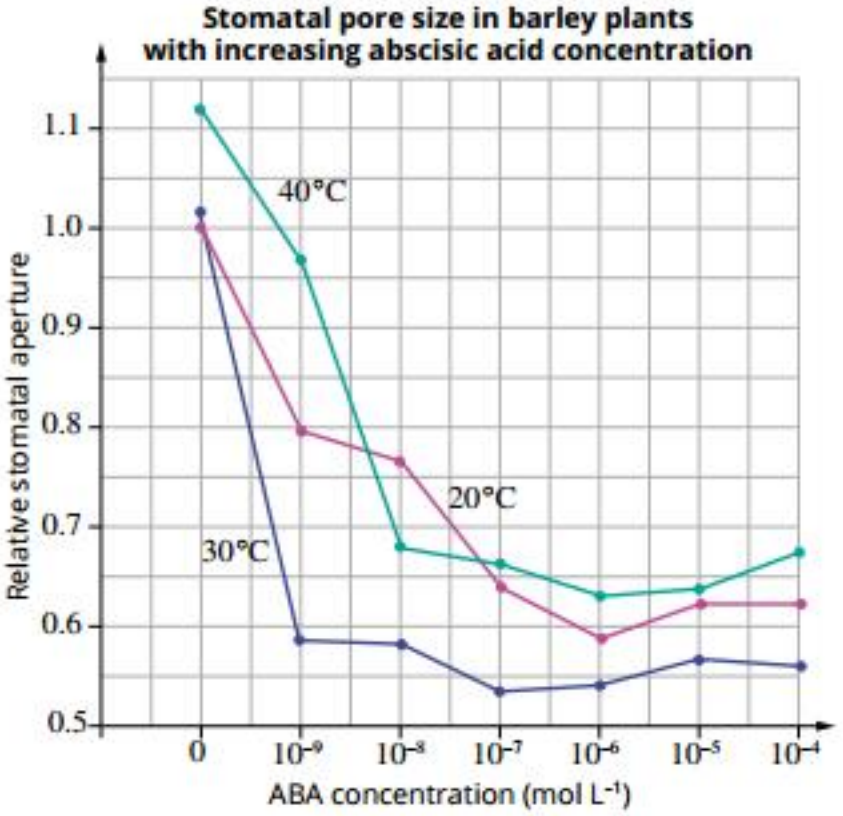
9 Guard cells, found in the leaf epidermis, are the entry point for carbon dioxide. Abscisic acid is a plant hormone that has been shown to be a significant contributor to guard cell opening and closing. When plants are under water stress guard cells lose turgor and close. As soils dry out, cells in the roots lose turgor. This stimulates these cells to produce abscisic acid.

a Explain how the abscisic acid produced in the roots is transported to the guard cells in the leaves.

b The diagram above shows three types of cell signalling in humans. Explain which type of signalling is most like the action of abscisic acid.



c The graph shows the size of the stomatal pore (relative stomatal aperture) of barley plants as the concentration of abscisic acid is increased. The experiment was undertaken at three different temperatures.



Interpret the information displayed on the graph to answer the following questions.

i At which temperature was stomatal aperture size most affected by an increase in abscisic acid?

ii Describe the trends in stomatal aperture size as abscisic acid concentration increases at 40°C.

Chapter review

06

KEY TERMS

abscisic acid (ABA)
action potential
adaptation
adrenaline
aestivation
afferent (sensory) neuron
aldosterone
antidiuretic hormone (ADH)
autonomic nervous system
auxin
axon
axon terminal
baroreceptor
carotid rete system
catecholamine
cell body
central nervous system (CNS)
chemoreceptor
cortisol
counter-current blood flow
cytokinin
dendrite
dopamine
ectotherm
effector
efferent (motor) neuron
eicosanoid
endocrine system
endotherm
ethylene
external environment
gibberellin
glucagon
glycogen
heat exchange
hibernation
homeostasis
hormone
hydrophyte
insulin
internal environment
interneuron
kleptothermy
mechanoreceptor
mesophyte
myelin sheath
negative feedback loop
nervous system
neuron
neurotransmitter
nociceptor
nodes of Ranvier
non-shivering thermogenesis
osmolality
osmoreceptor
oxytocin
peripheral nervous system (PNS)
photoreceptor
phytohormone
piloerection
polarised
positive feedback loop
receptor
reflex
renin
sensory neuron
shivering thermogenesis
signal transduction
soma
stomata (sing. stoma)
steroid
stimulus
synapse
synaptic terminal
thermoreceptor
thyroid hormone
transpiration
torpor
vascularised tissue
vasoconstriction
vasodilation
xerophyte

KEY QUESTIONS

Describe

1 Homeostasis in an organism is achieved by:
- **A** external conditions
- **B** enzymic reactions
- **C** mostly negative feedback loops
- **D** mostly positive feedback loops

2 Identify which of the ions listed below does not assist in the generation of an action potential.
- **A** Na^+
- **B** Li^+
- **C** Ca^{2+}
- **D** K^+

3 Gibberellins, cytokinins and auxins are all recognised examples of:
- **A** phytohormones
- **B** osmoregulation hormones
- **C** thermoregulation hormones
- **D** mammalian reproductive hormones

4 Antidiuretic hormone (ADH) is important in water regulation for humans. Another hormone that helps regulate water balance in humans is:
- **A** insulin
- **B** renin
- **C** thyroxine
- **D** thyroid-stimulating hormone

5 Recall which two systems are the most important in regulating the internal environment of animals.

6 List the three neuron types and distinguish the function of each.

7 Explain how structural and homeostatic mechanisms maintain water balance in plants, including the roles of stomata, vacuoles, cuticle and abscisic acid.

8 Describe how the location of stomata in plants can help to regulate water loss.

9 Describe how signal transduction occurs from one neuron to the next neuron.

10 Explain the process of evaporative heat loss in animals to regulate temperature and cool quickly. Use an example to support your explanation.

11 Explain the role of ADH in homeostasis.

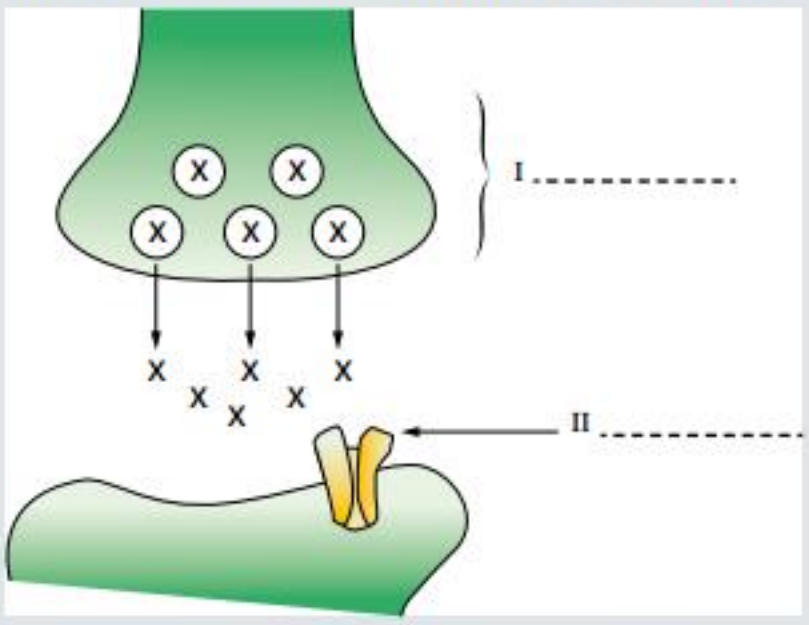
12 **a** Explain the principle of negative feedback in maintaining homeostasis.

b Using a diagram, explain how a decrease in body temperature can be reversed. In your diagram, draw and label an arrow to show where negative feedback occurs.

Apply

13 Thermoregulation is essential for the survival of endothermic organisms. One way the human body can generate heat is by piloerection. Describe this process and how it generates heat.

14 Identify and label parts I and II in the diagram below.



15 A plant is losing water from its tissues on a hot and windy day. What is this process called and what possible response could the plant undergo to prevent further water loss?

16 Discuss two responses that occur within the human body when blood glucose levels are above the normal range.

Analyse

17 An experiment was conducted to investigate the effects of changing gibberellin concentration on the germination of commercially available flowering plants. Five-hundred seeds were soaked in varying solutions of gibberellins as shown in the table below.

Gibberellin (mg/L)	Germination percentage after 20 days
0	9
10	20
50	21
100	22
200	33
500	55
1000	85

a State the function of gibberellin.

b What is the relationship between the concentration of gibberellin ($mg\,L^{-1}$) and germination?

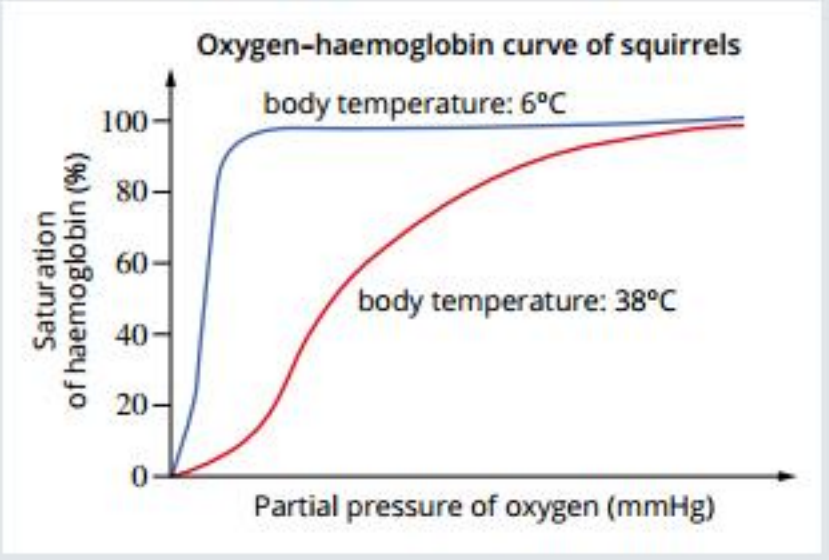
c The sensitivity of a flowering plant cell to gibberellin depends directly upon the number of receptors that the target cell has in its membrane for that hormone. Describe the relationship between number of hormone receptors and sensitivity to that hormone.

18 Cortisol is an important human hormone. It has a role in glucose regulation, immune system regulation and regulation of metabolic rate.

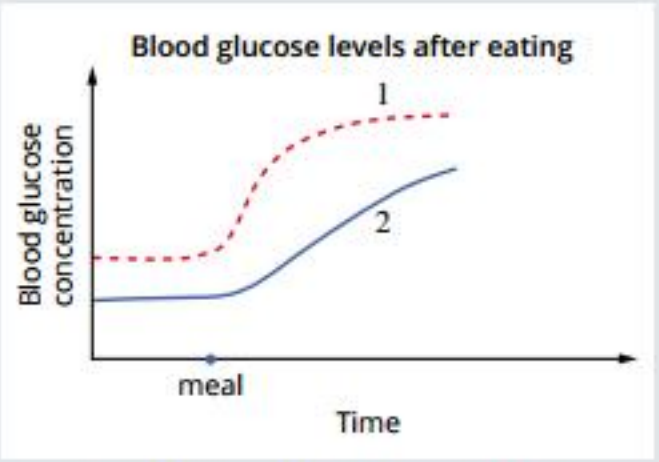
a Despite its role in many aspects of human physiology, not all cells respond to cortisol. Explain why some cells do not respond to cortisol.

b Insulin is the hormone that stimulates the uptake of glucose by cells. Fat and muscle cells are generally particularly sensitive to insulin, but cortisol is known to limit their response to this hormone. Infer how cortisol could reduce the normal response by fat and muscle cells.

19 During hibernation, the oxygen–haemoglobin dissociation curve of squirrels shifts to the left as shown on the graph below. This means that oxygen binds more readily to haemoglobin in the blood. Determine how this might be an advantage to a hibernating animal.



20 Blood glucose levels in individuals with and without diabetes, after eating similar meals are shown in the graph below.



- **a** Determine which results (1 or 2) displayed on the graph represents the individual with diabetes. Explain your reasoning.
- **b** Draw a diagram to represent the negative feedback model for the control of blood glucose levels in the body.

21 You will recall from Module 6.4 the physiological aspects of hibernation, which is a form of torpor. Mammals that hibernate are generally quite small (bats, rodents and pigmy possums) and yet bears (brown, black, grizzly or polar) are also said to be hibernators. A bear's body temperature may only drop a few degrees, compared to other hibernators whose temperatures can go down to as little as 5°C.

Based on what you know of torpor and hibernation, judge whether or not, bears are true hibernators.

22 The table below shows the rate and relative percentage of blood flow to the various organs and tissues of the body at rest and during strenuous exercise.

	Blood flow			
	Rest		**Exercise**	
	Amount ($mL min^{-1}$)	**Proportion of total (%)**	**Amount ($mL min^{-1}$)**	**Proportion of total (%)**
brain	750	13	750	5
heart	250	5	750	5
muscle	1200	20	12 500	71
skin	500	9	1900	11
kidney	1100	19	600	3
abdomen	1400	24	600	3
other	600	10	400	2
Total	5800		17 500	

- **a** Assess by what factor the blood flow rate increases from rest to exercise.
- **b** Distinguish which organs/tissues experience a decrease in blood supply from those that have an increase.
- **c** Determine why these changes occur.
- **d** Interpret from the data, which organs/tissues have the greatest overall increase or decrease in their blood supply.
- **e** Infer whether or not the data indicates that the demands of the brain are any different during exercise.

Interpret

23 The following question is based on your having completed Practical investigation 4. Ideally, you would have examined a range of plant leaves from open forest, grassland, aquatic systems and closed or rainforest plants to ascertain stomatal distribution. You would have carefully noted the distribution of the stomata for each type of plant and have devised an overall theory of stomatal distribution of Australian plants.

But you may not have encountered beaded samphire (*Sarcocornia quinqueflora*), which is a common succulent halophyte growing in salt marsh areas that are regularly inundated by tides along most of the Queensland coast.

- **a** Based on your experimental results, predict the stomatal and guard cell distribution of this plant.
- **b** Having made your prediction, list in order of preference (whether or not you have access to coastal regions), the steps you will take to validate your statement.

24 Experiments have shown that plants show positive phototropism as the result of the movement of auxin to the side of the shoot that receives less light. Devise experiments to demonstrate the following research statements and briefly list the procedures you would follow.

a Gravity is an important factor in the movement of auxin.

b The degree of bending of the shoot does not vary with the colour of light available.

Data analysis

DATA SET 1

The information below applies to Questions 1–5.
Research findings have indicated that low oestrogen levels are directly linked to osteoporosis (the loss of calcium from bone), causing a decrease in bone mass. Osteoporosis over time causes bones to become brittle and easier to damage. A study was conducted on 122 postmenopausal women. The women were randomly assigned to a treatment with either calcium supplement (1000 mg per day) or a placebo (no calcium supplement) for two years. The bone mineral density (BMD) of the total body was measured every six months. A bone mineral density (BMD) test measures calcium and other minerals in bone. Bones containing more minerals are denser, so they tend to be stronger and less likely to break. The results of the study are shown the graph below and are expressed as a percentage of the base-line values.

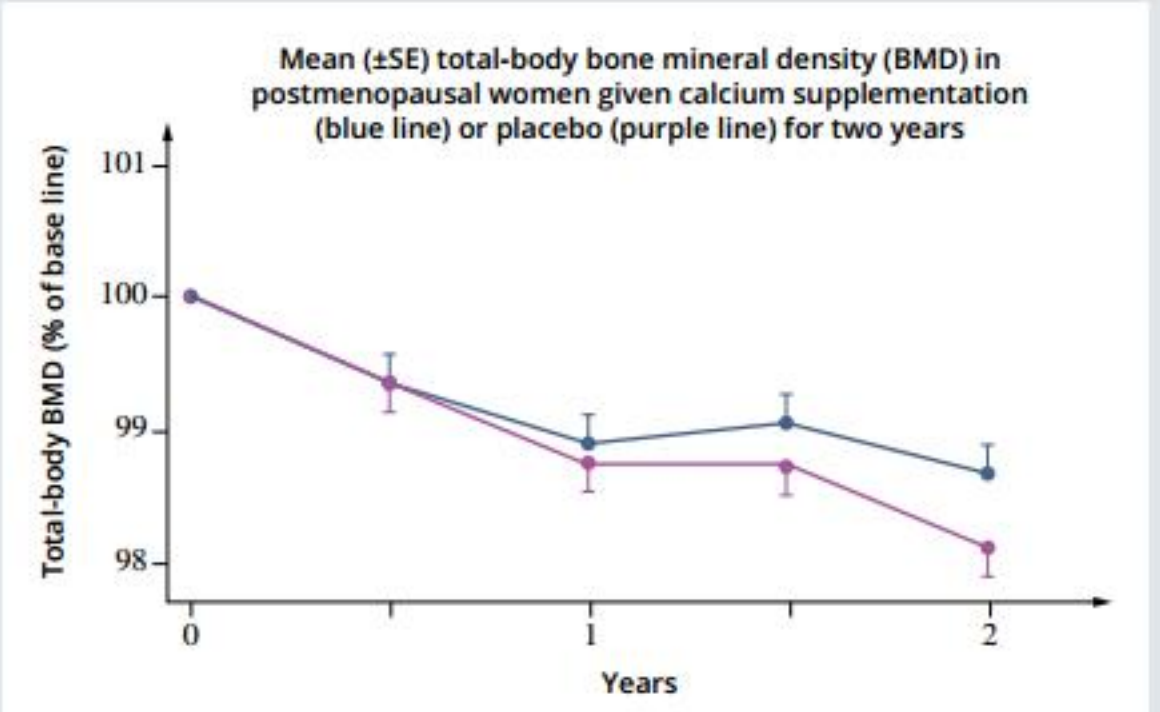


FIGURE 1 Bone mineral density (BMD) in postmenopausal women given calcium supplements over two years.

Question 1 (1 mark)

Identify when the greatest bone mineral density measurement was recorded.

Question 2 (1 mark)

What was the bone mineral density measurement after two years for the treatment group?

Question 3 (1 mark)

What was the bone mineral density measurement after two years for the placebo group?

Question 4 (2 marks)

Describe the relationship between bone mineral density and years in both the supplement and placebo patients.

Question 5 (1 mark)

What do these results indicate about the use the calcium supplements (taken postmenopausal)? Use data to support your answer.

CHAPTER

07 Disease and its causes

This chapter is about disease and its causes. You will learn about genetic, environmental and nutritional causes of non-infectious disease, as well as cellular and non-cellular pathogens associated with infectious disease in plants and animals and how they enter the host and cause harmful alterations to the functioning of the host's body. You will also examine data about the prevalence and cause of disease to learn to evaluate the accuracy, validity, and reliability of scientific data.

Syllabus subject matter

Topic 2 • Infectious disease and epidemiology

- Distinguish between infectious and non-infectious disease. **7.1**
- Identify key features of prions, viruses, bacteria, fungi, protists and parasites. **7.2, 7.3**
- Explain how adherence factors, invasion factors, capsules and toxins affect pathogenesis. **7.2**

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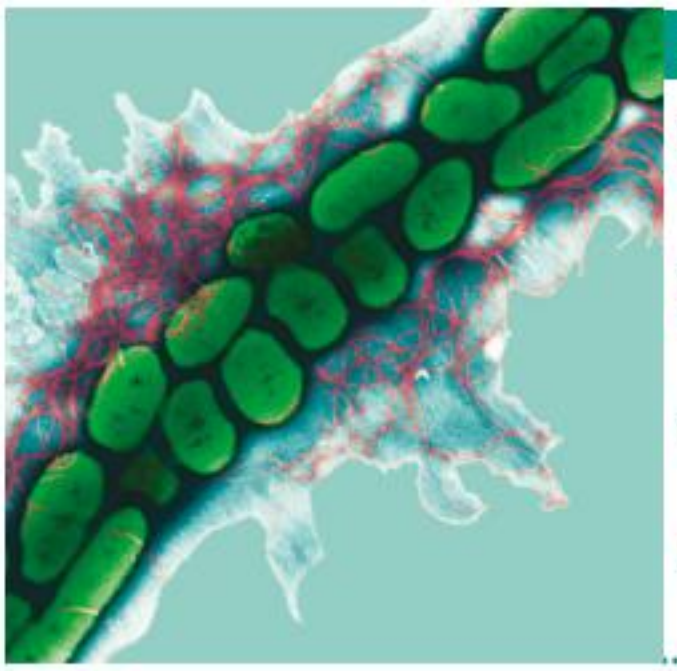
7.1 Non-infectious disease



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- describe the difference between infectious and non-infectious disease
- understand that non-infectious diseases can be caused by genetic, environmental and nutritional factors
- explain the concept of heterozygote advantage
- provide specific examples of genetic, environmental and nutritional diseases and their causes
- understand the usefulness of studies of fraternal and identical twins in determining the relative contributions of genetics and environment to the development of disease
- recognise the role of statistical analyses such as regression analysis in determining the cause of disease.

A **disease** is any condition that impairs the normal functioning of an organism. The symptoms of disease can result from tissue destruction, blocked transport systems, starvation, or malfunction due to an overuse of resources, the production of toxic substances or genetic mutations. Disease symptoms may also result from an undesirable immune response, such as an allergic reaction, or an attack on the organism's own cells as occurs with auto-immune diseases. Immune rejection, which causes severe disease, is a current hurdle that medical innovation is trying to overcome by tissue transplant and cellular therapies. You will recall learning about stem cell therapies in Chapter 4 Cell specialisation and organisation.

Many diseases are caused by **infectious** agents, called **pathogens**, which can be passed from one individual to another. Infection occurs when a microorganism becomes established and grows in a host, regardless of whether the host is harmed. Infection is not synonymous with disease because microorganisms growing inside a host do not always result in host damage, but when they do, this is referred to as an **infectious disease**. Infectious diseases are caused by pathogenic microbes such as bacteria, viruses, fungi, parasites or prions and can spread (i.e. be transmitted) from one individual to others.

Non-infectious diseases result from genetic, environmental or lifestyle factors. While the term 'transmitted' can be used in reference to genetic diseases, it is more precise to use the terms 'inherited' or 'passed down'. This is because genetic diseases are passed from parents to their offspring through genes, and 'transmission' is more commonly associated with infectious disease.

Non-infectious diseases fall into three broad categories.

- Inherited diseases are passed down through genes during reproduction.
- Nutritional diseases are caused by insufficient or inadequate diets.
- Environmental diseases are the result of environmental factors, such as exposure to dangerous chemicals.

Many non-infectious diseases result from a combination of genetic, environmental and nutritional factors, rather than a single cause. In addition, lifestyle factors such as physical activity, smoking, alcohol consumption and stress can also influence the risk of developing non-infectious diseases. For example, asthma results from a complex mix of genetic, environmental and lifestyle factors. Genetic predisposition plays a significant role in the development of asthma, which can be both caused and triggered by environmental exposure to irritants, pollutants or allergens. The condition is also more common in people who are classified as obese due to mechanical effects on lung function and obesity-related inflammation.

Another example is cancer, which is characterised by the uncontrolled division of cells that can spread and invade surrounding tissues. There are many types of cancer and the type someone has depends on the specific cells that become cancerous. Disease symptoms vary based on the type of cancer someone has, how far it has spread and what tissues and organ systems are impacted.

Mesothelioma (a type of lung cancer) has been linked to environmental exposure to asbestos. The chemicals in cigarette smoke significantly increase the risk of many cancers, including throat and lung cancer. Excess exposure to ultraviolet radiation causes skin cancers such as melanoma (Figure 7.1.1). Several strains of the human papilloma virus (HPV) can increase the risk of developing cervical cancer. Evidence also shows that nutritional deficiencies in iron, zinc, folate and vitamins B6, B12 and C, can cause DNA damage that increases cancer risk.

FIGURE 7.1.1 Melanoma is a type of skin cancer. Key features are irregular borders and colouration, asymmetry, a raised surface compared with the rest of the skin, and a diameter that is typically greater than 6 mm.

Genetics plays a strong role in the development of cancer. People can have a lower or increased risk of cancer because of their genetic make-up. For example, two genes that are active in breast tissues are *BRCA1* and *BRCA2*. The normal versions of these genes are involved in regulating cell division in healthy breast tissue. Mutations in these genes have been linked to a loss of cell division regulation and subsequent increased risk of developing breast cancer. Mutations in *BRCA1* and *BRCA2* have also been linked to an increased risk of developing ovarian cancer.

The risks of developing breast or ovarian cancer are listed in Table 7.1.1.

TABLE 7.1.1 Risk of developing breast or ovarian cancer for people with and without *BRCA* mutations who have not previously been diagnosed with either of these cancers

	Incidence in the general population (%)	Incidence in individuals with mutated *BRCA1* or *BRCA2* gene (%)
female breast cancer by age 70	7	56–87
ovarian cancer by age 70	<2	27–44
male breast cancer by age 70	0.05	6

GENETIC DISEASES

All cells contain DNA. This molecule contains the information needed to build all of the cells that make up the organism. The DNA that carries the information for making a protein, such as an enzyme, is called a gene. The DNA of organisms is arranged into 'packets' called **chromosomes**. Humans have 46 chromosomes in all of their body cells and 23 chromosomes in their gametes (sperm and ova).

When new cells are made, the chromosomes are copied so that each of the new cells has a complete copy of the DNA. On rare occasions, during the formation of gametes, the DNA is copied incorrectly, or the gamete ends up with too many or too few chromosomes. This is called a **mutation**. Some mutations can be inherited by future generations while others can be fatal or allow the individual to survive but render them unable to reproduce.

Some mutations always result in disease; others do not directly cause disease but increase an individual's chances of developing a specific condition. Many types of cancer, type 2 diabetes and heart disease show this pattern.

Chromosomal diseases

Most chromosomal diseases occur when too many chromosomes end up in a gamete and thus in the individual. The cause of such diseases is identified by photographing the person's chromosomes and arranging them in pairs. The photograph of a person's chromosomal pairs is called a **karyotype**. Using the karyotype, it is possible to determine whether a person has extra or missing chromosomes in their cells, or if there are extra or missing pieces of individual chromosomes. Disease in these cases is caused by having too much or not enough of the proteins coded for by the genes on that chromosome. A common condition caused by too many chromosomes is Down syndrome (trisomy 21), which is the result of having a third copy of the number 21 chromosome (Figure 7.1.2).

Single gene diseases

In sexually reproducing organisms, such as humans, half of all the DNA of an offspring is inherited from each parent. Each parent provides half of the information needed for each trait. The interaction between these two pieces of information, called **alleles**, determines the trait of the offspring. Normally, inheritance of a mutated allele does not cause disease because the other allele inherited from the other parent provides sufficient information to make the necessary protein. People with two different alleles of a particular gene are described as being **heterozygous** for that gene. If the two alleles of an individual gene are the same, the person is **homozygous** for that gene.

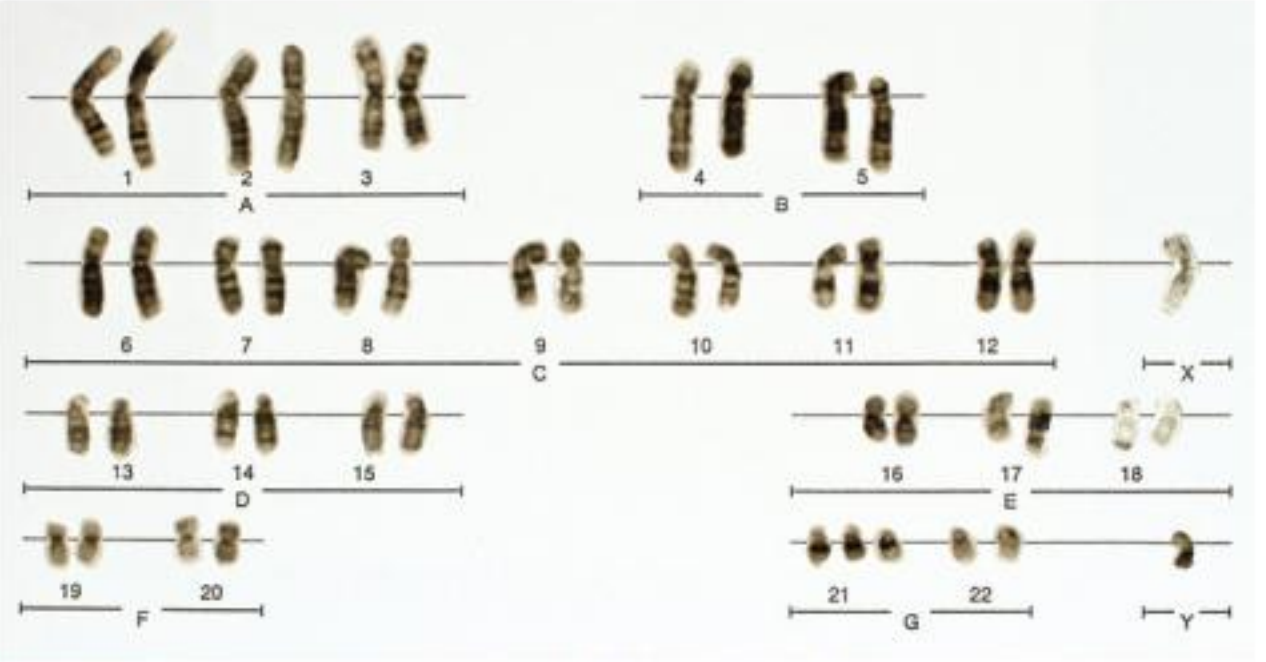


FIGURE 7.1.2 A karyotype of a human male with Down syndrome. Instead of two members of the 21st pair, there are three.

If a person inherits mutated alleles from both parents, then the required proteins are not made, or they are made but do not work properly, which can result in genetic disease. The only possible cure for a genetic disease is genetic engineering, which is highly experimental and only being tried in a few specific cases, but treatments are available for many genetic conditions.

There are many inherited diseases. These may have only minimal impact (e.g. red–green colour blindness) or they may have very serious consequences (e.g. sickle–cell anaemia and cystic fibrosis).

Sickle–cell anaemia

Sickle-cell anaemia is a condition in which red blood cells do not form correctly (i.e. they are malformed). Normal red blood cells are soft and flexible, enabling them to move easily through blood vessels. By comparison, the malformed red blood cells (sickle cells) that cause sickle–cell anaemia have a crescent shape (Figure 7.1.3), which makes them 'sticky' and as a result they become stuck in small blood vessels, blocking the flow of blood and causing symptoms typical of the disease.

The function of red blood cells is to carry oxygen around the body. To do this, red blood cells contain two types of haemoglobin, alpha and beta, which are proteins (see Module 5.2). Beta haemoglobin is coded for by the *Hb* gene. People with sickle-cell anaemia have a variant of this gene, called *HbS*. In order to have sickle-cell anaemia, a person must inherit *HbS* from both parents (Figure 7.1.4).

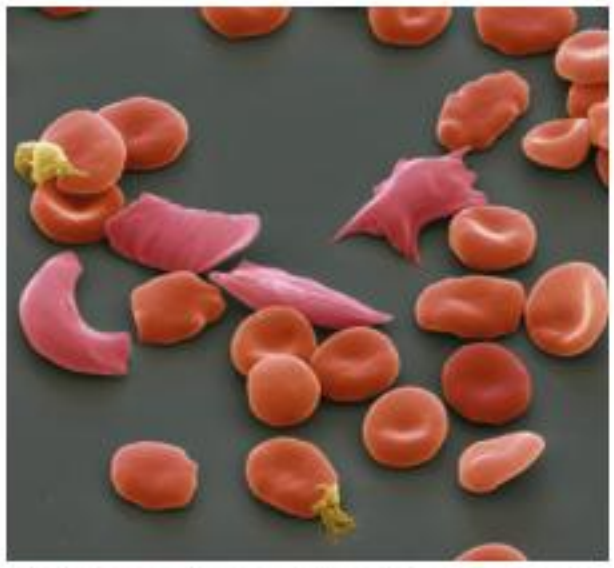


FIGURE 7.1.3 A scanning electron micrograph of red blood cells of a person with sickle-cell anaemia. The sickle-shaped red blood cells (pink) become stuck in small blood vessels, causing blockages that result in the symptoms of the disease.

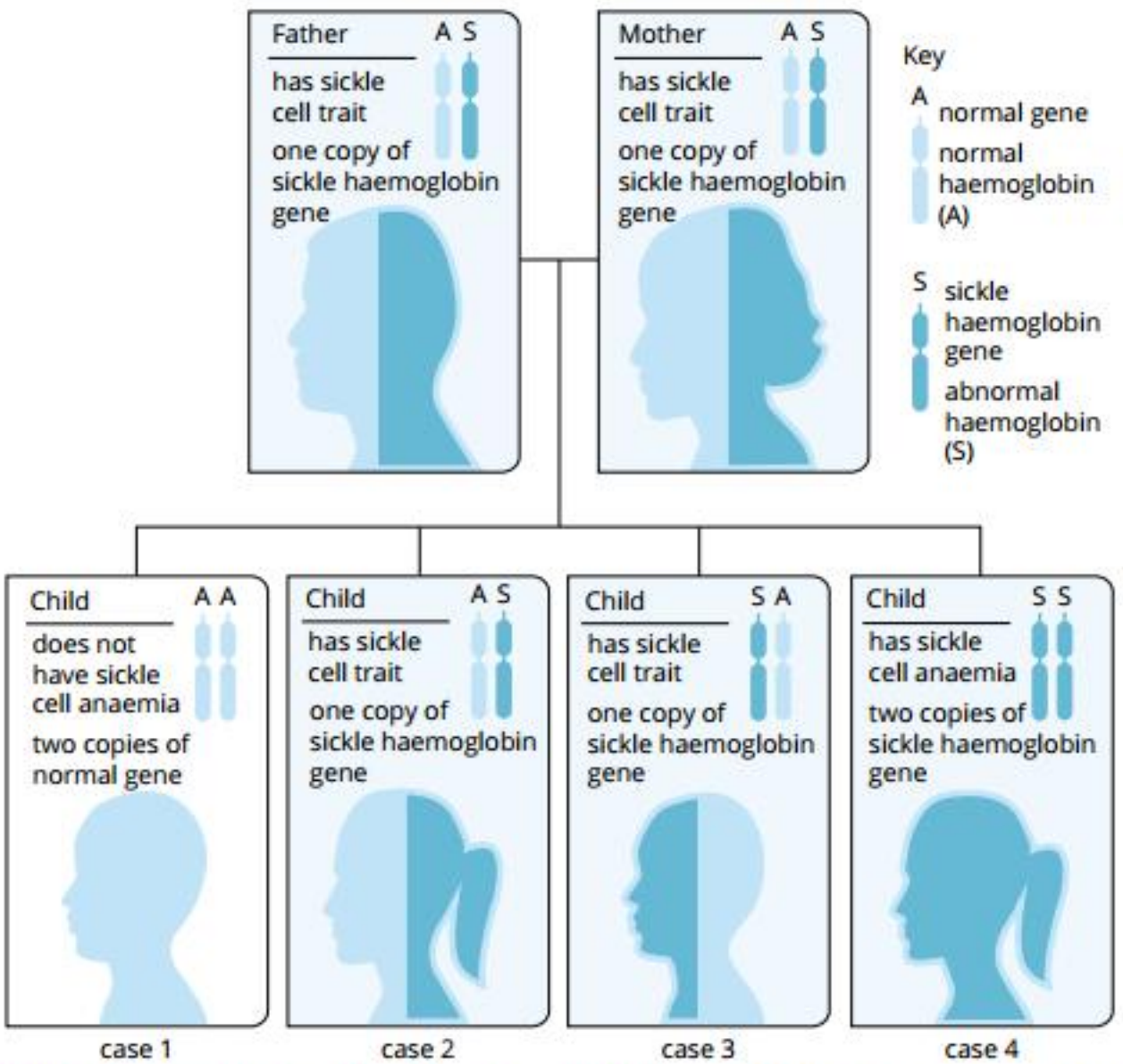


FIGURE 7.1.4 A couple who both have the sickle-cell trait have a one in four chance of having a child with sickle-cell anaemia.

People who are homozygous for sickle-cell anaemia—that is, they inherit the trait from both parents—suffer continuous problems, including severe joint pain, swelling of hands and feet, vision problems due to damage of small blood vessels in the eyes and frequent infections. They also have a significantly increased risk of stroke caused by blockages in the blood vessels in the brain.

Unlike normal red blood cells, which live for around 120 days, sickle cells survive for only around 20 days. The short life span of sickle-cell red blood cells means there is a severe shortage of red blood cells, so people with sickle cells have a lack of oxygen to tissues and experience extreme tiredness.

People who are heterozygous for sickle-cell anaemia—that is, they inherit the trait from only one parent—have the sickle-cell trait. These people generally live normal lives because under normal conditions their red blood cells are normal (biconcave) and they have no symptoms. However, the red blood cells of people who are heterozygous for sickle-cell anaemia may deform to a sickle shape when their body is under extreme stress such as during severe dehydration or severe illness, as occurs when the malarial parasite infects the person.

In studying the prevalence of the disease sickle-cell anaemia, epidemiologists noticed that the frequency of the disease seemed to align with the frequency of malaria caused by a protozoan, *Plasmodium falciparum*. Further research showed the ability of *P. falciparum* to enter red blood cells, where they spend a part of their life cycle, was severely reduced when the red blood cells were sickle-shaped. This led to the hypothesis that having the sickle trait helped people to resist malarial infection but did not significantly reduce their lifespan. Their cells only became sickle-shaped during infection. *P. falciparum* could not enter the cells to hide from the immune system and continue their life cycle and the people's red blood cells returned to normal once the pathogen was cleared. The protective value of having the sickling trait is thought to explain the high prevalence of the *HbS* allele in high malaria areas (Figure 7.1.5).

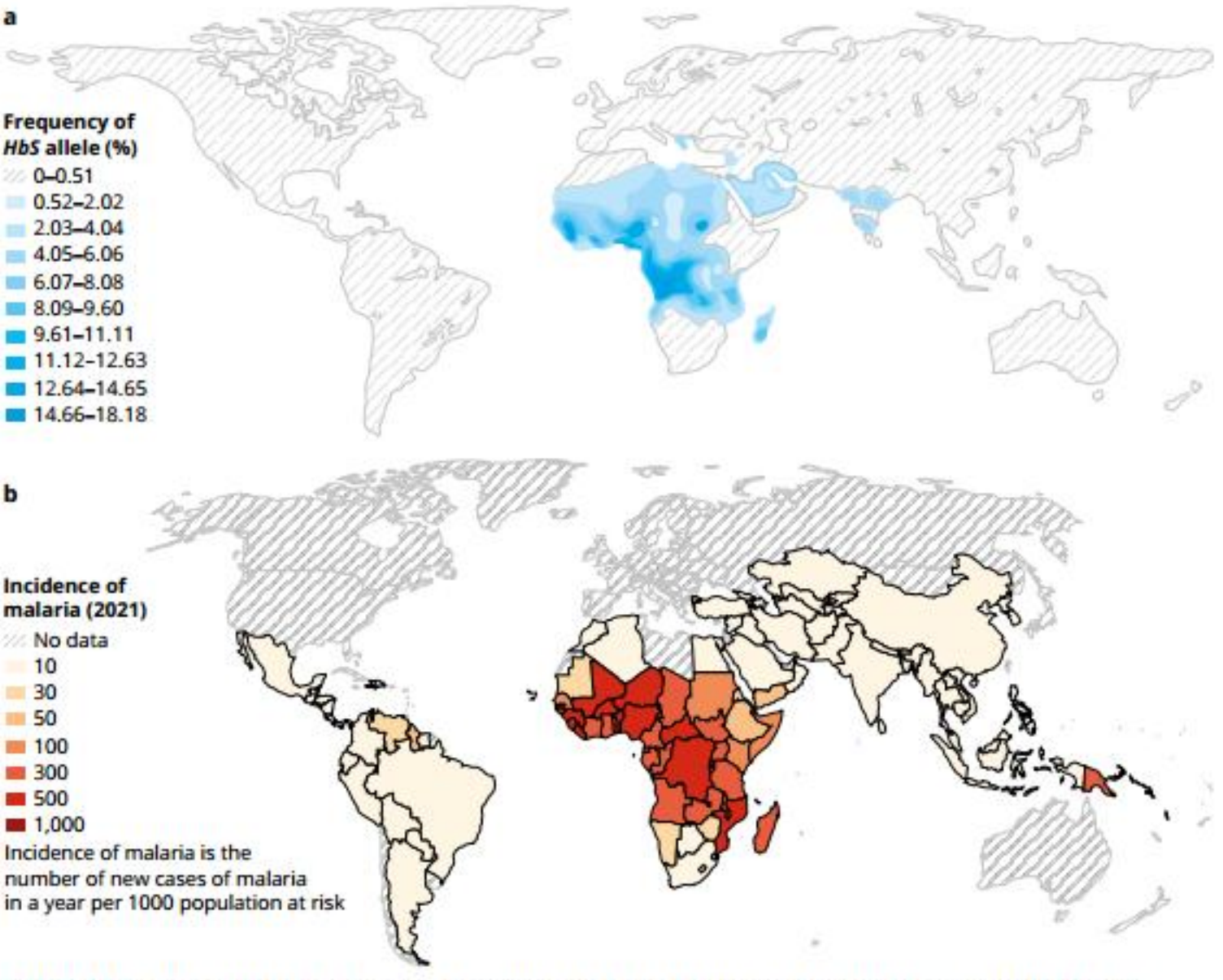


FIGURE 7.1.5 (a) Areas in Africa having high levels of malaria have a high frequency of the *HbS* allele. (b) The highest frequencies of the *HbS* allele, of 12.64% or more, occur in holoendemic areas. In holoendemic areas, virtually every individual is infected, childhood mortality is high, but most adults are largely asymptomatic due to having a level of tolerance.

Although the sickle-cell trait protects against malaria, it is not common in Asia and the Americas, where malaria rates are also relatively high. This is thought to be due to other similar genetically determined blood diseases in these areas, such as β-thalassaemia, which occurs around the Mediterranean and in Asia. Also, in Asia and the Americas, the most common cause of malaria is *Plasmodium vivax*, not *Plasmodium falciparum*, the latter of which is associated with higher rates of mortality. Thus, the advantage provided by the sickle-cell trait is greater in *P. falciparum* areas than in areas where *P. vivax* is more common.

Heterozygote advantage

Some genetic diseases are very low in frequency while others are relatively common, particularly in specific populations. This has resulted in scientists hypothesising that certain genetic traits, which can cause disease when inherited from both parents, convey protection against pathogens: this is called **heterozygote advantage**. These genetic diseases include cystic fibrosis (which may protect against typhoid, cholera, amoebic dysentery), thalassaemia (which may protect against malaria and coronary heart disease) and connexin–26 deafness (which may protect against bacterial ear infection). However, other scientists dispute that these mutations give increased resistance to disease.

For example, the heterozygote advantage conferred by cystic fibrosis was examined using a mouse model. Cystic fibrosis is a disease in humans caused by defects in the proteins that move salt across cell membranes. It results in the formation of thick, sticky mucus in the lungs, digestive tract and reproductive tract.

Mice with a mutation that caused the same reduction in the movement of salt and water across cell membranes as human cystic fibrosis were infected with cholera. The mice with the cystic fibrosis mutation suffered less dehydration than the mice without the mutation. This supported the hypothesis that cystic fibrosis genes also provide resistance to the effects of cholera. This would be an advantage to heterozygotes.

Many disease experiments are done using non-human animals as the subjects. However, non-human animals do not always respond as humans do and so any findings must be treated with caution when extrapolating to humans. A human study was done using data collected during a cholera **epidemic**. The severity of symptoms and rates of mortality were noted at the time of the epidemic, and tissue samples were analysed after the epidemic to determine whether individuals were heterozygous for cystic fibrosis. The data showed no difference in the severity of cholera between people heterozygous for cystic fibrosis and people who did not have the allele, so this study did not support the hypothesis that cystic fibrosis genes provide a heterozygote advantage to cholera.

Genetics, environment and disease

In many cases, diseases are not caused by a single genetic mutation. Instead, individuals show a genetic predisposition for a disease and the environment provides the trigger, which results in the disease manifesting. A genetic predisposition means that a particular individual has an increased chance of developing a particular condition. This is the result of having certain genetic variations. These variations are often inherited but may also result from new mutations. Having a genetic predisposition does not mean an individual will develop a disease, only that they are more likely to under certain conditions than a person without the predisposition.

It is often difficult to identify the relative influence of the genetic and environmental factors that contribute to disease. Despite having a genetic predisposition, disease may not develop in the absence of an environmental trigger being encountered by an individual. Diseases known to have a mixture of environmental and genetic causes are heart disease, many forms of cancer, asthma, obesity and type 2 diabetes.

Identifying the influence of genetics on disease—twin studies

With most forms of disease, genetics plays a role in determining either the chances of developing the disease or how severe the disease is if it occurs. One important method that scientists use to identify the role of genetics is twin studies.

There are two types of twins: identical and fraternal. Following fertilisation, the zygote starts dividing to grow into a new individual. Sometimes, for unknown reasons, the bundle of cells split (usually between the morula and late blastocyst stage) so that two separate bundles of cells are formed from the original fertilised egg. Both bundles grow to form a baby, and because both came from the same zygote, they are genetically identical. These are identical twins and are always the same sex.

Fraternal twins, or non-identical twins, arise when a female releases two eggs at the same time, both of which are fertilised by two different sperm. Both zygotes undergo division and become babies, but like all brothers and sisters, they arise from separate eggs and sperm from the same parents. They are no more genetically alike than any other siblings from the same parents and can be the same sex or different sexes.

It is useful for researchers to study identical and fraternal twins because there are fewer environmental differences to account for given that (1) twins are the same age and (2) developmental factors relating to age can be discounted as the cause of any difference between them. Studies of identical twins in particular can be used to identify environmental effects because identical twins have no genetic differences, so any difference in disease occurrence between the pair must be due to environmental factors.

The results of one such twin study looking at the causes of asthma are shown in Table 7.1.2. Of particular significance were the identical twins where only one twin had asthma. For females, 185 out of 1232 (or 14.94%) of the twin pairs had asthma. This is significant because it suggests there must be a substantial environmental component to the disease. The data for this study was collected through a survey sent to the participants who then self-reported whether they had ever had asthma. The results of surveys, especially those that involve self-reporting, can be less reliable because the researcher must rely on the participants to accurately assess and report on their own situation.

TABLE 7.1.2 Results of a study into asthma prevalence among adult twins

Type and sexes of twins	Total number of pairs who participated	Number of pairs with asthma (and percentage)			
		Both Yes	Yes No (elder twin Yes)	No Yes (elder twin No)	Both No
Identical, both female	1232	67 (5.4%)	87 (7.1%)	98 (7.9%)	980 (79.5%)
Identical, both male	567	39 (6.9%)	33 (5.8%)	31 (5.5%)	464 (81.8%)
Fraternal, both female	751	19 (2.5%)	67 (8.9%)	69 (9.2%)	596 (79.4%)
Fraternal, both male	352	12 (3.4%)	43 (12.2%)	39 (11.1%)	258 (73.3%)
Fraternal, male and female	906	32 (3.5%)	105 (11.6%)	98 (10.8%)	671 (74.1%)

Examining the differences between fraternal and identical twins can also indicate the importance of genetics in susceptibility to disease. For example, significant differences in disease prevalence between male and female twins could indicate a hormonal cause for the disease. However, if there is a difference in disease prevalence between first-born and second-born twins, then it may be that factors during the birthing process are a cause of disease (e.g. inhalation of amniotic fluid or increased stress due to a longer time birthing).

NUTRITIONAL DISEASES

Nutritional diseases include conditions that are caused by either inadequate or excessive intake of nutrients and/or kilojoules. In developed countries, many nutritional diseases are caused by excessive kilojoule intake and may involve insufficient intake of required nutrients such as vitamins and minerals. In developing countries, many nutritional diseases are associated with inadequate intake of both kilojoules and nutrients.

Lack of a vital component in the diet can cause **malnutrition** and deficiency diseases. For example, lack of vitamin C causes scurvy (Figure 7.1.6), lack of iron causes anaemia and lack of protein causes kwashiorkor (Figure 7.1.7). Starvation causes muscle and tissue wasting and a decreased metabolic rate.

FIGURE 7.1.6 Mild cases of scurvy, such as that shown here, are easily treated with increased vitamin C intake. Severe scurvy used to occur on long sea voyages. Severe scurvy results in bleeding into the joints, causing severe pain, loss of teeth, increased bruising and poor wound healing, shortness of breath and finally death.

Vitamin D deficiency

Aside from iron and vitamin D deficiency, nutritional deficiencies are uncommon in Australia, except in some Indigenous communities. While diet can be a source of vitamin D, most of our vitamin D is produced by skin cells when they are exposed to sunlight. Vitamin D deficiency has become more common as more Australians practice sun safety to reduce their risk of skin cancer, and is most common in southern areas of Australia, especially in the winter months when there is less sunlight. Relative risks of vitamin D deficiency in Australia for summer and winter are shown in Figure 7.1.8. Even in Queensland, 15 out of every 100 people experience vitamin D deficiency during winter.

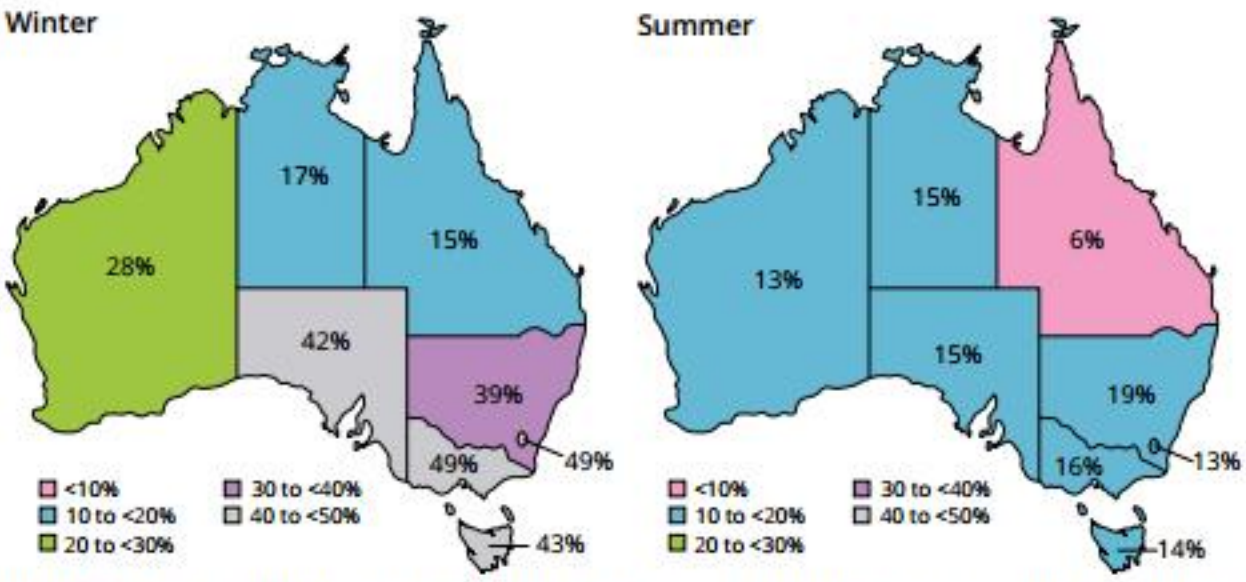


FIGURE 7.1.8 Maps of Australia showing the prevalence of vitamin D deficiency across the country in winter and summer. The map shows that there are significant levels of vitamin D deficiency throughout the year, even though Australia has high levels of ultraviolet radiation especially in summer.

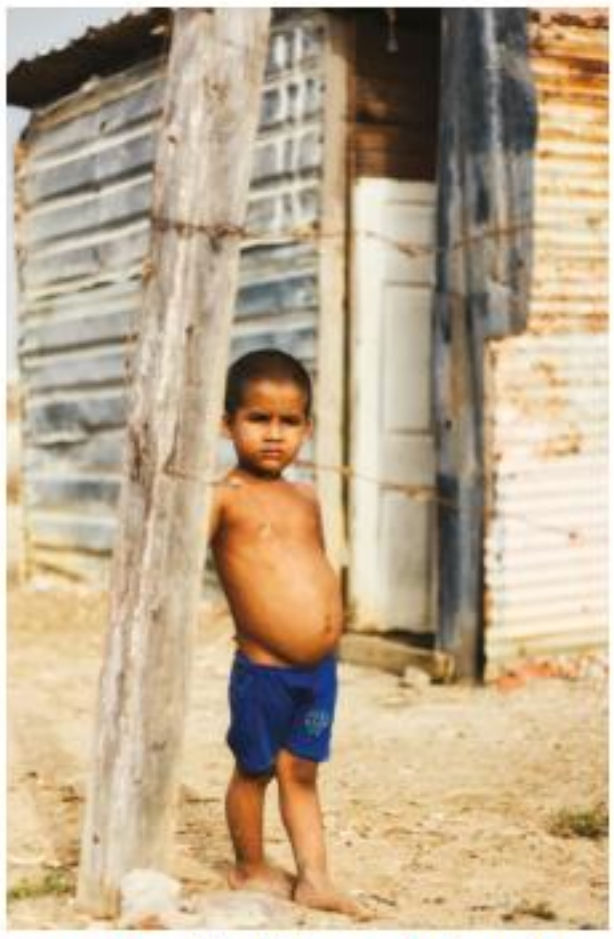


FIGURE 7.1.7 Kwashiorkor results in symptoms such as anaemia, tissue wastage, skin sores and fluid accumulation in the tissues (oedema), which results in a 'pot' belly.

Vitamin D is essential for building and maintaining healthy teeth and bones. Children who are vitamin D deficient are in danger of developing rickets. Rickets is a disease in which the bones become chalky and brittle. In adults, the most common consequence of vitamin D deficiency is osteoporosis or the less severe osteopenia, both of which are the result of low bone density. It is estimated there are more than 6.2 million Australians over the age of 50 with either osteoporosis or osteopenia. This will lead to more people requiring access to the health system for the treatment of broken bones.

Obesity

A common nutritional disease in Australia is obesity. Obesity occurs when the intake of kilojoules exceeds the energy used for daily activity over a substantial period. The Australian Institute of Health and Welfare reported that in 2017–18, 67% of adults and 25% of children aged between 2 and 17 were overweight or obese.

Hypervitaminosis is disease caused by excessive vitamin intake. It is most commonly associated with the fat-soluble vitamins (A, D, E, and K) because they are stored and accumulate in the body's fatty tissues and liver.

Being overweight has been linked to the increased risk of several life–threatening conditions, including:

- type 2 diabetes
- high blood pressure (hypertension)
- heart disease
- stroke
- fatty liver disease
- kidney disease
- sleep apnoea
- some cancers.

Links between some of these conditions and obesity have been identified through epidemiological studies that compare people with and without obesity, as shown in Figure 7.1.9.

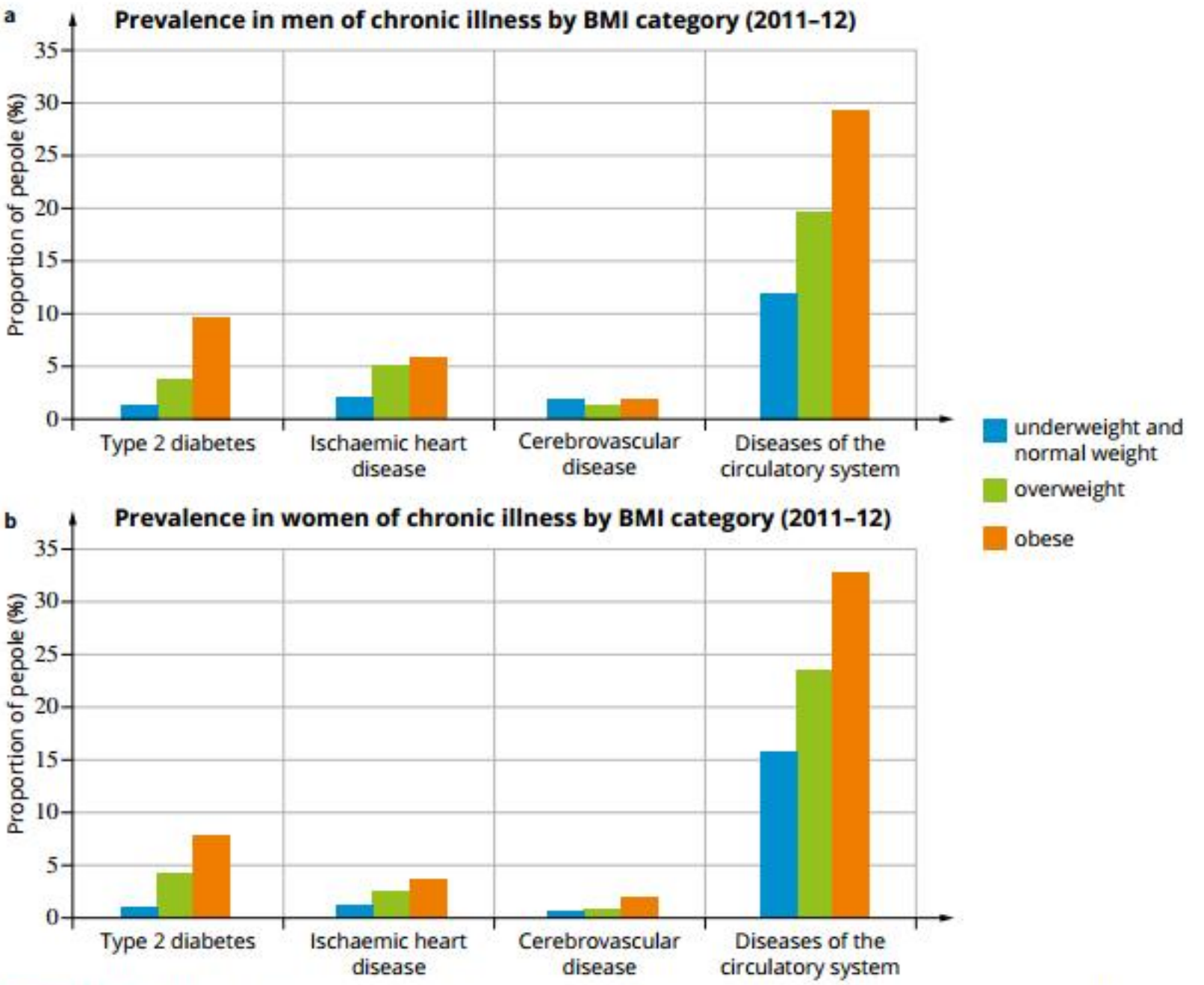


FIGURE 7.1.9 Prevalence of chronic illness by body mass index (BMI) category (2011–12) for (a) men and (b) women. The graphs indicate that higher weights lead to higher levels of disease. This study was undertaken by the self-reporting method.

The link between hypertension and obesity required long-term studies of people and the collection of large datasets. Comparisons were then made between groups with and without obesity and prevalence was calculated. In the case of obesity, type 2 diabetes and cardiovascular disease as shown in Figure 7.1.9 (cardiovascular disease includes ischemic heart disease, cerebrovascular disease and diseases of the circulatory system), a significant difference in prevalence of type 2 diabetes and cardiovascular disease was observed between people with and without obesity. The data indicates that type 2 diabetes and cardiovascular disease were more prevalent in obese men and women compared to people who are not obese or who are underweight (Figure 7.1.9).

ENVIRONMENTAL DISEASES

Environmental diseases are caused by factors in the environment such as exposure to toxic chemicals, radiation, stress and pollutants. Early studies of disease by the scientists Robert Koch, Ignaz Semmelweis and others established that specific pathogens were causative agents of specific diseases. Today, **epidemiology** takes a broader approach and investigates the distribution, occurrence and determinants of health and diseases in specific populations. It includes the statistical analysis of data with the aim to identify cause-and-effect relationships of a disease. For example, the relationship between smoking and lung cancer was established using epidemiological studies.

CASE STUDY 7.1.1

Using statistical evidence to establish causal links for lung cancer

Today around 1 billion people smoke cigarettes, about half of whom live in China, and worldwide over 8 million people die from smoking-related diseases every year. About 50% of those deaths are from lung cancer. In Australia, smoking-related lung cancer kills about 7500 people each year.

The now accepted link between smoking and lung cancer was not firmly established until around 1950, even though suspicions arose early in the 20th century. The first suggestion that smoking tobacco was a cause of lung cancer appeared in scientific literature in 1912. By the 1930s, scientists and doctors were becoming more concerned about the rising incidence of lung cancers and noted that the use of cigarettes was also increasing (Figure 7.1.10), and during the 1940s and 1950s, epidemiological studies and evidence from animal experiments provided strong evidence of the link between smoking and lung cancer.

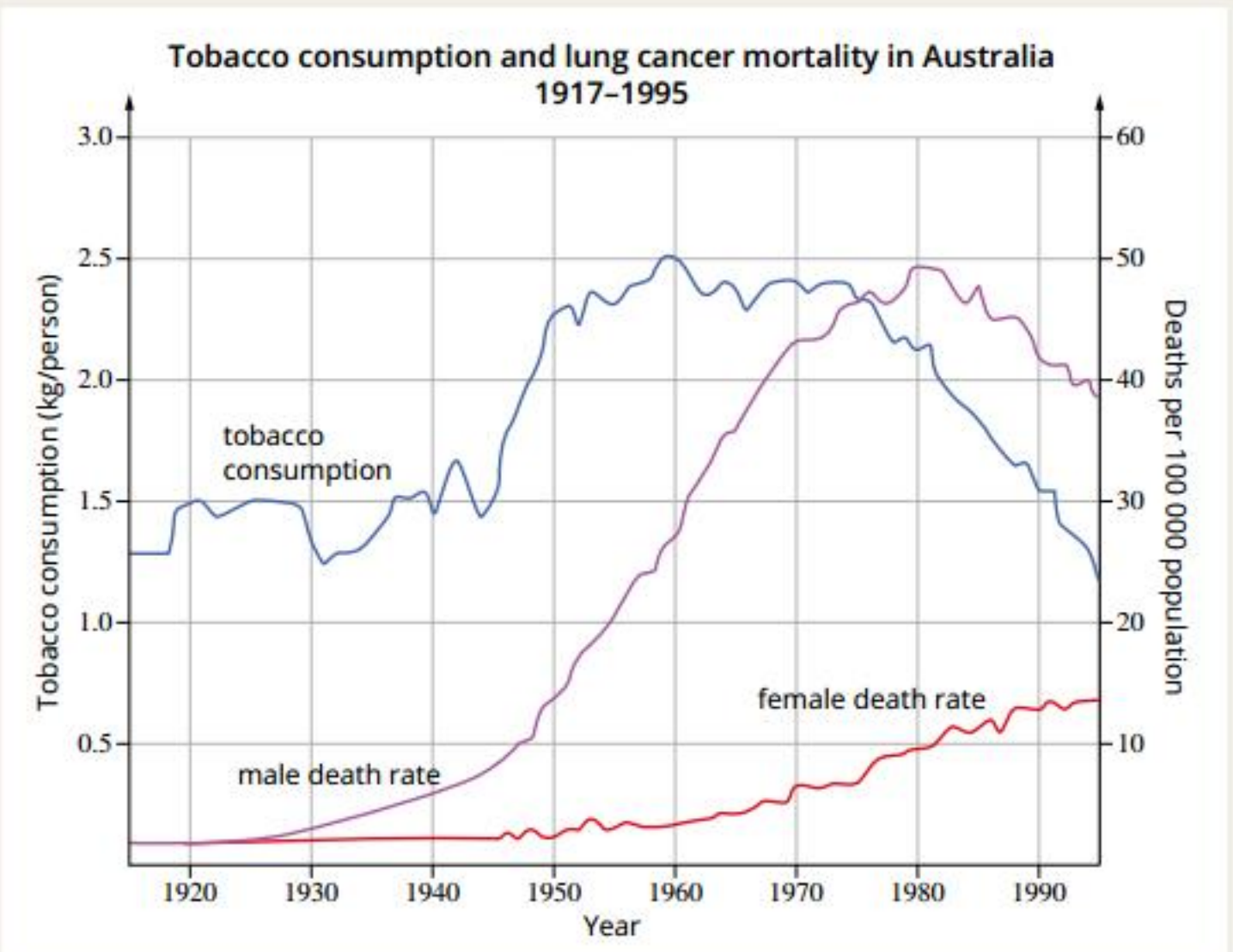


FIGURE 7.1.10 Tobacco consumption and lung cancer mortality in Australia between 1917 and 1995. Shortly after tobacco consumption declined, so did death rates.

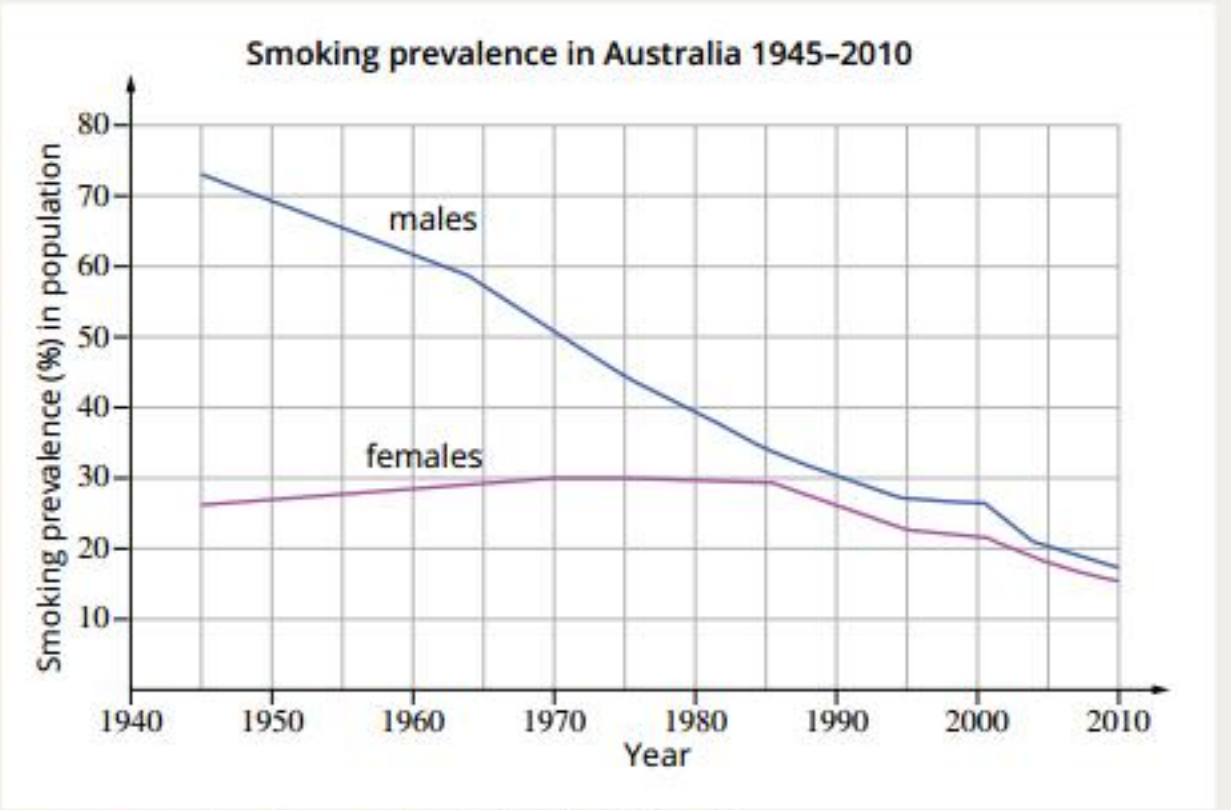


FIGURE 7.1.11 Smoking prevalence in Australia 1945–2010

The time lag between the reduction in smoking and reduction in cancers (Figure 7.1.10) is now attributed to the time needed for the carcinogenic properties of tobacco to build up to the point of causing disease. However, the time taken for tobacco to cause cancer meant that the link was hard for many people to accept. Acceptance of the link between smoking and cancer was made even more difficult by the tobacco industry publishing studies arguing there was no causal link between smoking and lung cancer. Despite this, by the mid-1960s people in developed countries began to accept the relationship between smoking and lung cancer. Subsequently, smoking and cancer rates began to fall, especially among men.

The decline in women's lung cancer rates took longer as smoking prevalence peaked and began to decline among women much later than for men (Figure 7.1.11). Statistics for 2014–15 show that smoking has declined even further to 16.9% of men and 12.1% of women, so Australia can expect a continuing decline in the rates of lung cancer caused by smoking.

Review

1 Identify the decade in which female smoking began to decline.

2 Discuss why epidemiological studies were needed to establish the link between smoking and lung cancer.

3 Compare the declining trend of tobacco consumption and male death rates.

Other epidemiological studies have established causal links between:

- leukaemia and exposure to benzene
- heart attacks and high cholesterol
- salt intake and hypertension
- lead and brain damage.

Epidemiological studies can also show that, in some instances, there are no causal links between environmental factors and disease. For example, a few years ago there was a media campaign by a group who opposed vaccinations. They suggested that the MMR (measles, mumps and rubella) vaccine caused autism. Large-scale studies were undertaken that showed that there is no evidence of a causal link between the MMR vaccine and autism.

CASE STUDY 7.1.2

Does early exposure to lead cause life-altering brain damage?

Investigation into some environmental diseases requires long-term studies to produce results. A number of long-term studies have been undertaken on the effects of lead exposure on children. It has long been known that lead is toxic; writings as early as 250 BCE mention gastric problems and anaemia caused by lead. We now also know that lead damages the nervous system and especially the brains of children.

In the human population, the mean intelligence quotient (IQ) is 100, with a standard deviation of 15 and 95.44% of all scores falling within two standard deviations of the mean. For many years, arguments occurred about the possibility that exposure to lead caused brain damage, especially in children, and that this damage would affect IQ.

A large number of studies in various countries indicated that all levels of exposure to lead are harmful. One particular series of studies, involving cities in a number of countries (including Port Pirie, South Australia, and Mount Isa, Queensland) investigated the consequences of lead exposure in young children. The studies specifically looked at the effect of lead exposure on brain damage, using IQ deficit as a measure of damage to the brain.

These studies indicated that any level of lead is harmful, but that severe damage begins at 5 $\mu g\,dL^{-1}$ of blood, increases rapidly up to 10 $\mu g\,dL^{-1}$ and then decreases slowly thereafter.

It was established that there is a step-like pattern to the damage associated with lead-caused brain damage. The critical points seemed to be 5 $\mu g\,dL^{-1}$ and 10 $\mu g\,dL^{-1}$ (Figure 7.1.12).

Within each graph, the trendline and measure of variance (R^2) shows that there is very little significance to any variations within each section. This shows that within these groups, there is not much change in IQ deficits. However, examining the means of each group shows a significant trend (Table 7.1.3 and Figure 7.1.13).

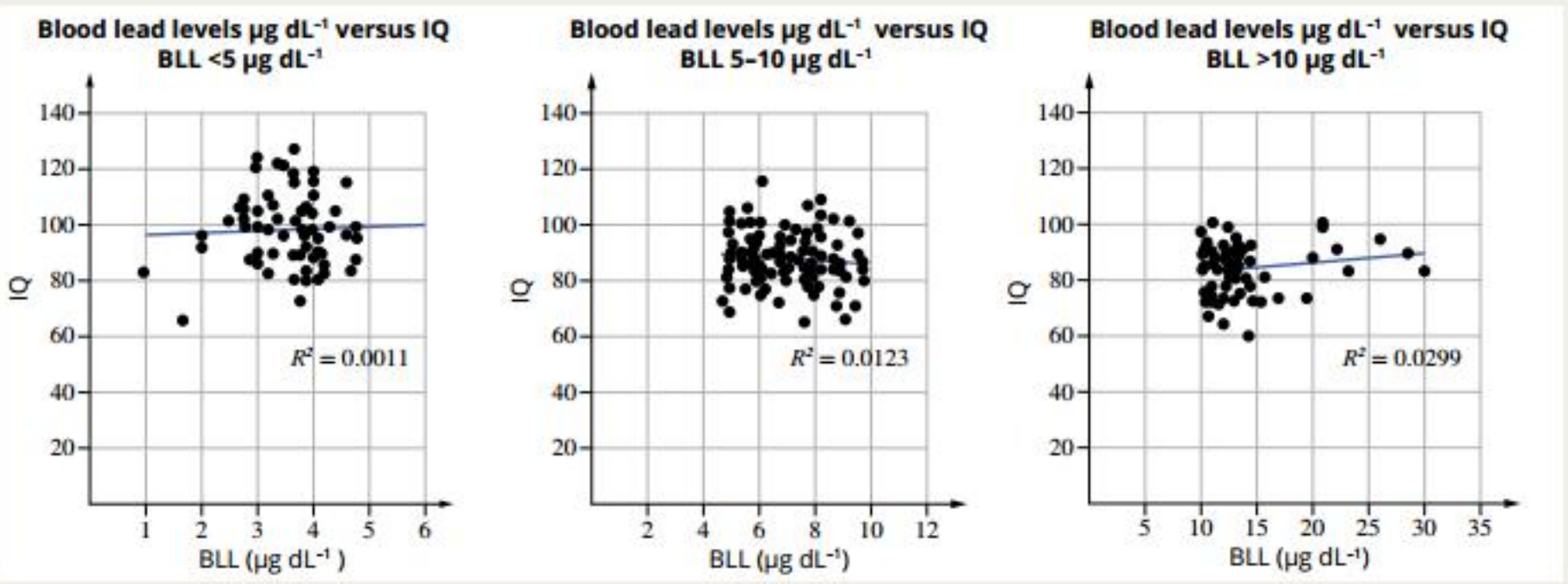


FIGURE 7.1.12 Blood lead levels (BLL) versus IQ. The data collected from one study is represented in the three scatterplots. Trend lines and R^2 values were calculated using Excel

1 µg is one-millionth of a gram and 1 dL is one-tenth of a litre.

TABLE 7.1.3 Relationship between mean blood lead level and mean IQ

Blood lead level ($\mu g\,dL^{-1}$)	Mean blood lead level ($\mu g\,dL^{-1}$)	Mean IQ	Standard deviation for IQ	Number of subjects
<5	3.6583	98.49	12.64	71
5–10	7.076	89.111	9.2109	116
>10	13.668	84.103	9.054	67

Most studies showed similar results, which indicated that mean IQs of children exposed to lead had an average IQ deficit between 10 and 13 points even once other factors such as health and socio-economic status were considered. Most of the studies took place in mining towns where there was a perceived problem, but the results of some of the earlier investigations correlated with changes in society. Beginning in 1985, leaded petrol was phased out in Australia because of concerns about the effects of lead on brain development.

Some scientists are now looking at lead-caused brain damage and correlating it with crime statistics. In many countries, there seems to be a correlation between falling levels of childhood exposure to lead (due to the reduction in the use of leaded petrol) and falling levels of crime as those children reach adulthood. As the graph in Figure 7.1.13 shows, there is a strong correlation between the two factors. However, such correlations must be treated with caution. Correlation does not equal causation. The possibility of coincidence must be ruled out, as well as the possibility that the two factors are related because they have the same cause. One factor that could cause both high-blood-lead in children and a high crime rate 23 years later is growing up in an economically disadvantaged area. In such areas, schools are often of a lower standard and children suffer educational and social disadvantage, which increases the risk they will commit crimes later in life. Also, in those areas the housing stock tends to be older and more poorly maintained. Older houses are more likely to contain lead paint, which was used in the past. If the houses are poorly maintained, such paint would be flaking from the walls, which would increase lead exposure. In carrying out the studies, the researchers had to ensure they were not just studying the effects of living in an underprivileged area.

Review

1. Compare the spread of data in Figure 7.1.12 for the different BLLs and the typical population IQ results of 100 with a standard deviation (σ) of 15 and 95.44% within 2σ.
2. The ancient Romans undertook many advanced engineering projects including the piping of water into cities and even individual wealthy homes. Unfortunately, they used lead pipes for most of this movement of water and some historians think that this may have contributed to the fall of the Roman Empire. Discuss the plausibility of this hypothesis.

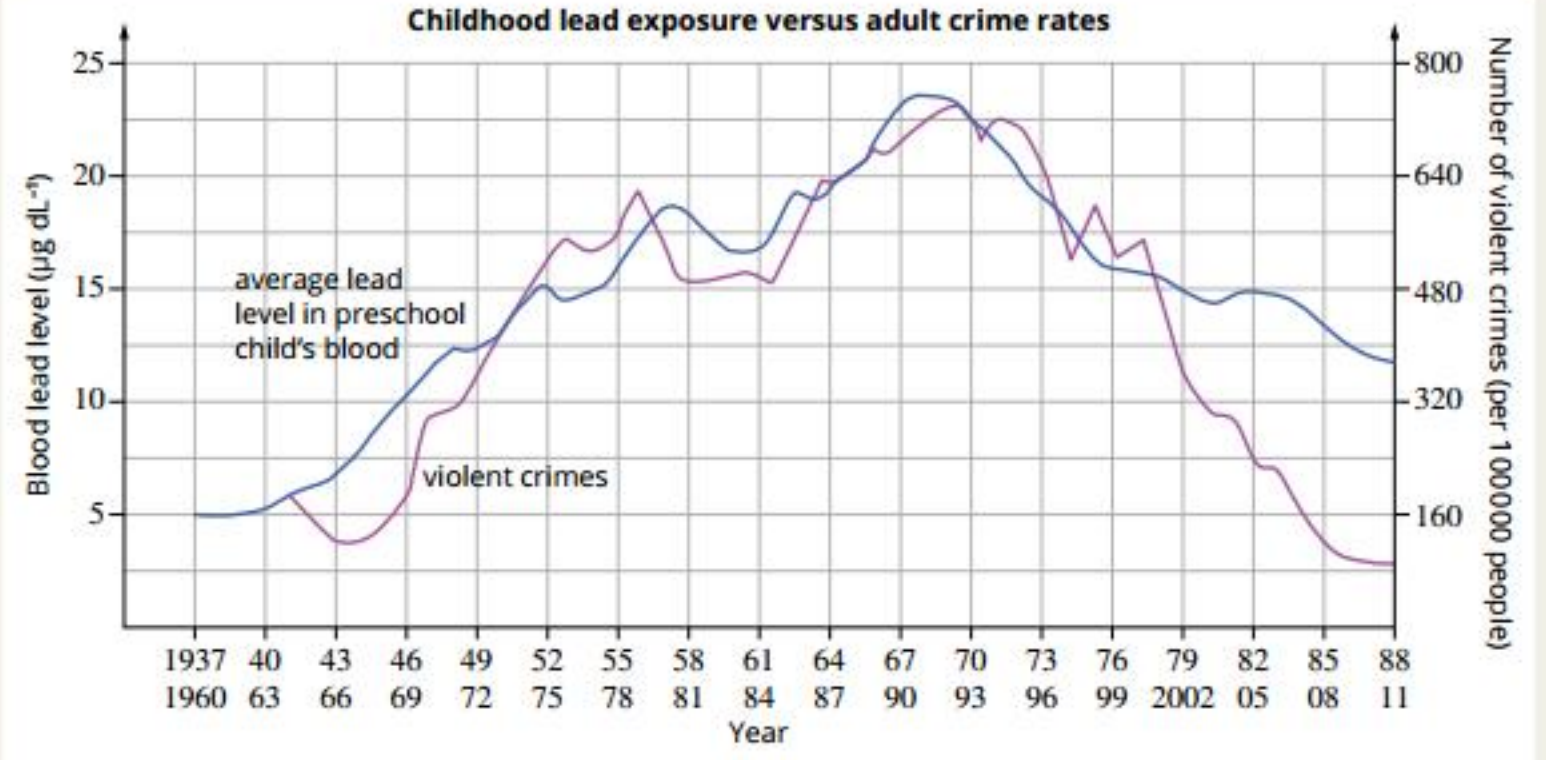

Childhood lead exposure versus adult crime rates
Blood lead level (μg dL⁻¹)
Number of violent crimes (per 100000 people)
average lead level in preschool child's blood
violent crimes
25
20
15
10
5
800
640
480
320
160
1937 40 43 46 49 52 55 58 61 64 67 70 73 76 79 82 85 88
1960 63 66 69 72 75 78 81 84 87 90 93 96 99 2002 05 08 11
Year

FIGURE 7.1.13 A comparison between childhood lead exposure (1937–1988) and adult crime rates (1960–2011) seems to show a strong correlation.

7.1 Review

SUMMARY

- Disease is any disruption to normal body functioning.
- Diseases can be caused by pathogens, environmental factors, nutritional factors, genetics or combinations of two or more of these factors.
- Genetic diseases are caused by mutations (changes) in genes or having too many or too few chromosomes.
- In the case of some single gene diseases, heterozygotes have an advantage in resisting certain pathogens; for example, sickle-cell anaemia and malaria.
- Nutritional diseases include conditions that are caused by either inadequate or excessive intake of nutrients and/or kilojoules.
- Large-scale studies, and studies of fraternal and identical twins assist in determining the relative contributions of genetics and environment to the development of disease.
- Individuals may have a greater or lesser chance of developing cancer depending on their genes and the environment to which they are exposed.
- Both smoking cigarettes and childhood exposure to second-hand smoke are significant environmental causes of disease.

KEY QUESTIONS

Describe

1 List three broad categories of non-infectious disease and state the main cause of each of them.

2 Name two diseases caused by nutritional deficiencies. State the specific cause and describe the symptoms of each disease.

3 Identify the dietary components that are lacking in Kwashiorkor and anaemia.

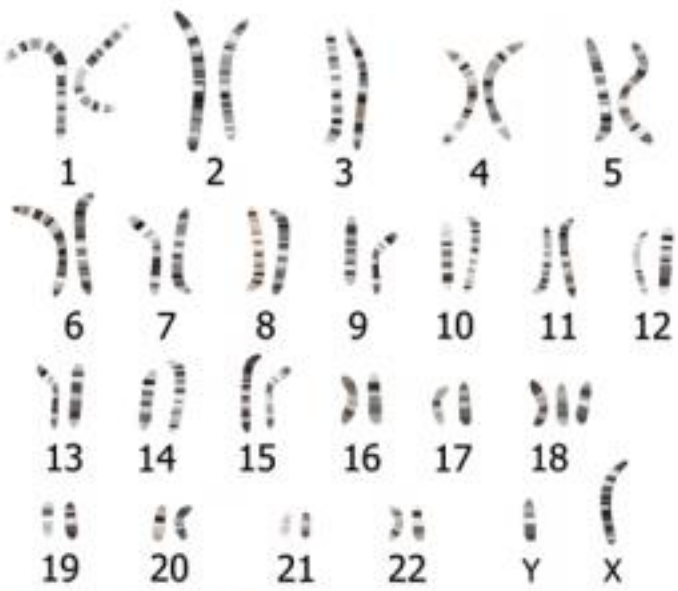
4 Why does the inheritance of a mutated allele not always cause disease?

5 What is the difference between infectious disease and non-infectious disease?

Apply

6 Cancer always requires a genetic predisposition to develop. Explain why this statement is incorrect.

7 Examine the following image.



a Name this type of image.

b Identify the abnormality shown in the image.

8 **a** Explain why establishing the cause of many non-infectious diseases involves the collection of very large amounts of data.

b Explain why twin studies are so useful in separating the genetic and environmental components of disease.

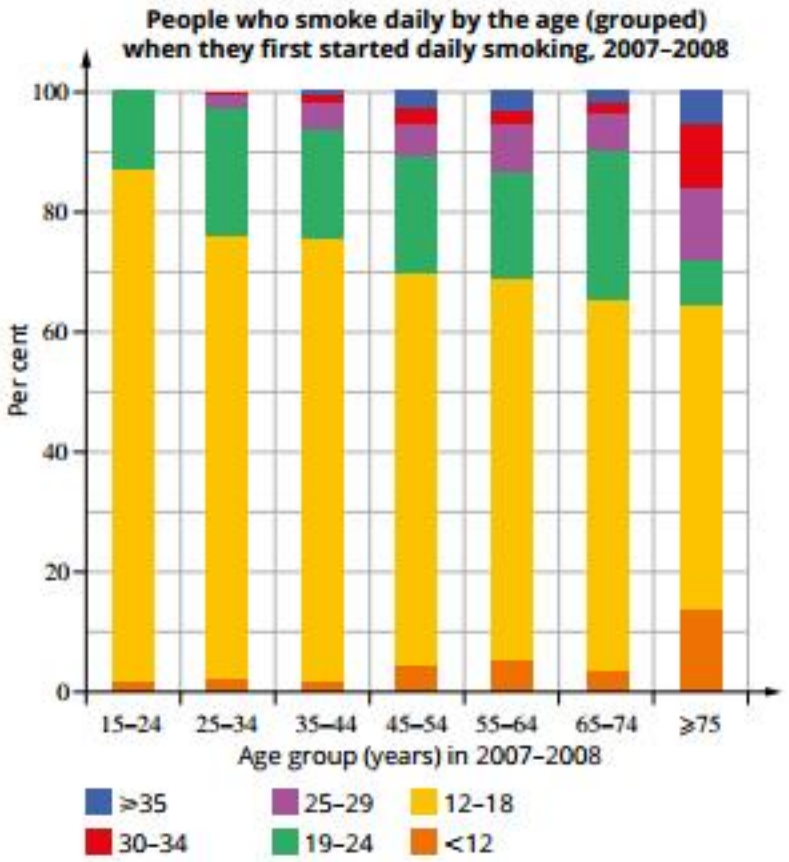
9 Identify the type of non-infectious disease that results from vitamin D deficiency and justify your choice.

continued over page

7.1 Review *continued*

Analyse

10 Smoking is a major contributor to the development of non-infectious diseases such as lung cancer and heart disease. For this reason, the Australian Health Survey collects data on the prevalence of smoking in the Australian population. The following graph shows the number of Australians in 2007–08 who smoked daily in age groups and the age when they first started to smoke (colour coded).



a Identify the age group from the period 2007–08 that contains the highest percentage of people who took up smoking before their teens.

b Determine the percentage of 35–44 year olds who took up smoking when they were between 19 and 24 years.

c **i** Calculate for each age group, the approximate percentage of people who took up smoking before their 25th birthday.

ii In a population of smokers, determine the likelihood that you will meet someone who started to smoke after the age of 25 years.

d Conclude what the graph indicates about age of uptake of smoking for the age groups 18 years and younger over the last several decades. Specify the evidence you used for your conclusion.

e Propose a significant piece of data that cannot be determined from the graph.

11 In a number of countries, including Australia, there have been numerous studies of the effect of childhood exposure to lead and the incidence of violent criminal behaviour later in life. One such study examined people living in an Australian city. This was a longitudinal study—children's blood lead levels were measured and then 20 years later the incidence of violent crime in the area was recorded. The data collected in the 2007–08 survey is summarised in the following table.

Year 1	Air lead levels ($\mu g\,m^{-3}$)	20 years later	Assault rate per 100 000 people
1991	0.39	2011	97.5
1989	0.73	2009	112
1988	0.84	2008	112
1990	0.78	2010	120
1987	0.78	2007	123
1986	0.92	2006	125
1985	0.99	2005	133
1983	1.10	2003	137.5
1984	1.22	2004	142.5
1981	1.30	2001	138
1980	1.30	2000	143
1982	1.39	2002	148

a Draw a graph in which you plot air lead levels against assault rate per 100 000 people.

b Draw a linear regression line and calculate R^2. Explain whether the value for R^2 suggests that there is a correlation between lead exposure and violent crime 20 years later.

c **i** Propose other factors that could account for a decrease in assault rates from 1982 to 1991.

ii Explain why other factors should be considered.

d Propose other information that would be valuable to increase confidence in the conclusion that exposure to lead in childhood results in higher levels of violence in adults.

12 Obesity has been established as a contributor to a large number of health problems. Study of the dietary behaviour of obese people and those with a healthy weight has significant problems because it is difficult to gain accurate information about what individual people eat. Generally, three sorts of studies are used:

- a food diary in which people write down what they eat each day at a time convenient to them
- a 24-hour retrospective report in which people are interviewed and report on what they ate the previous day
- a food frequency questionnaire in which the subject is given a long list of foods, and they tick off what they ate over the period of the study, which could be a day, a week, a month or even a year.

The reliability of these studies has been questioned. Discuss the sources of error for each type of study. In your discussion, propose the effect of the errors on the accuracy of the evidence and identify which type of study is likely to be the most reliable.

13 The following table displays the body mass index (BMI) of five individuals.

a Independently undertake research online using trusted sources to determine what BMI thresholds represent an increased and higher risk of type 2 diabetes. Explain why the source/s used can be trusted.

b Referring to the data in the table below, identify the individual/s who are:

i at an increased risk of developing type 2 diabetes

ii at highest risk of developing type 2 diabetes.

Individual	BMI ($kg\,m^{-2}$)
1	21
2	28
3	36
4	25
5	30

14 The following table shows the relationship between high blood pressure, deaths due to all cardiovascular diseases, and obesity rates, by state and territory.

a Calculate regression analyses of the following using the data in the table:

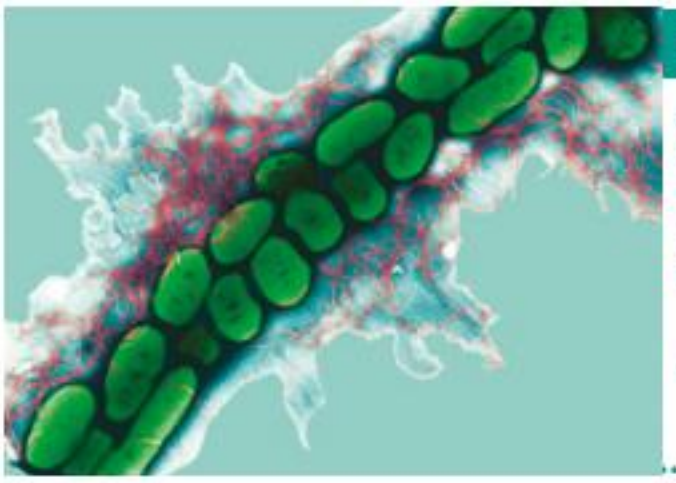
- obesity rates/high blood pressure by state/territory
- obesity rates/% deaths due to all cardiovascular diseases
- high blood pressure by state/territory/% deaths due to all cardiovascular diseases.

State or territory	High blood pressure by state/territory (%)	Deaths due to all cardiovascular diseases (%)	Obesity rates
Queensland	22.5	30.6	30.2
Tasmania	25.2	30.5	32.3
South Australia	22.6	30.2	30.0
Victoria	22.8	29.3	26.4
New South Wales	21.3	30.7	28.2
Western Australia	20.4	27.1	24.6
ACT	23.7	28.4	29.0
Northern Territory	21.5	22.5	23.9

Source: Australian Bureau of Statistics—2014/15 Australian Health Survey

b Explain what the R^2 values indicate about the relationships between the three factors.

7.2 Cellular pathogens



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that pathogens are a major cause of infectious disease
- recognise different types of cellular pathogens
- identify possible methods of disease spread
- understand how the interaction between a pathogen and the host determines the severity of the disease
- explain how evidence is used to support theories in regard to disease incidence and spread.

PATHOGENS AND PATHOGENESIS

Pathogens are agents that cause disease. Many diseases are infectious; that is, they are caused by pathogens that can be passed from one organism to another. These pathogens may be cellular or non-cellular.

Depending on their ability to cause disease, pathogens are divided into two groups.

- Primary pathogens cause disease any time they are present.
- Opportunistic pathogens only cause disease when the host's defences have been weakened. For example, by poor nutrition, stress, or infection by other pathogens.

Most cellular pathogens are **parasites**. They complete part of their life cycle in or on another organism, known as the host. All parasites live at the expense of their host, but some cause no significant harm or disease in normally healthy hosts. Sometimes, the parasite infects two different hosts in sequence—the **primary host** is used for the adult stage of the parasite and the **intermediate host** is used for the larval stage. **Endoparasites** (e.g. tapeworms) live inside an organism; **ectoparasites** (e.g. lice) attach to the outside.

Some organisms are not true parasites but they can transmit pathogens between hosts, such as the plague (or Black Death), carried by fleas, and malaria and zika virus, carried by mosquitoes. Organisms that carry pathogens between organisms are called **vectors**.

An infectious disease that spreads from non-human animals to humans and vice versa is called a **zoonotic disease** (or zoonosis). It has been estimated that 60% of current infectious diseases of humans are zoonotic and that 75% of emerging diseases started out in non-human animals. Infection by pathogens can occur through direct contact such as by touch; by contamination from faeces; by inhaling droplets of moisture released during breathing, coughing, and sneezing; through exchange of body fluids; and through cuts and scratches (see Chapter 9 Disease Patterns for more details).

When infection occurs through direct contact, the disease is said to be **contagious**. Highly contagious diseases such as influenza can spread rapidly through unprotected populations (see Chapter 9 Disease Patterns).

Infection can also occur by injection with a contaminated needle, or by bites from insects and other vectors. Another important source of infection is ingestion of contaminated food or water (see the case studies in Chapter 9 Disease Patterns).

The potential ability of a pathogen to cause disease in a host is called its **pathogenicity**; not to be confused with **virulence**, which is the severity of disease caused by a pathogen. Traits of both the host and pathogen combine to determine a pathogen's virulence. For example, some pathogens do not always infect the same tissues. Which tissues are infected can have a significant influence on the severity of the disease, as in the case of *Bacillus anthracis*, which causes anthrax.

B. anthracis can infect the skin, where it causes lesions that, with antibiotics, usually heal. However, if *B. anthracis* infects the lungs it is fatal in more than 80% of cases.

Characteristics of the host, such as its level of resistance to infection and the speed and strength of its immune response, will also affect the virulence of a pathogen.

After infection has occurred and the pathogen has become established, there is a period during which the pathogen reproduces and multiplies. During this period, the host may not show any signs of disease. Once the incubation period is completed, most hosts will show symptoms of disease. Sometimes, though, the host does not become symptomatic even while they remain infectious, in which case they are known as a **carrier**. Carriers are an important method of spreading disease.

The period between infection and the onset of symptoms is called the **incubation period**. Figure 7.2.1 shows the progress of disease from infection to wellness.

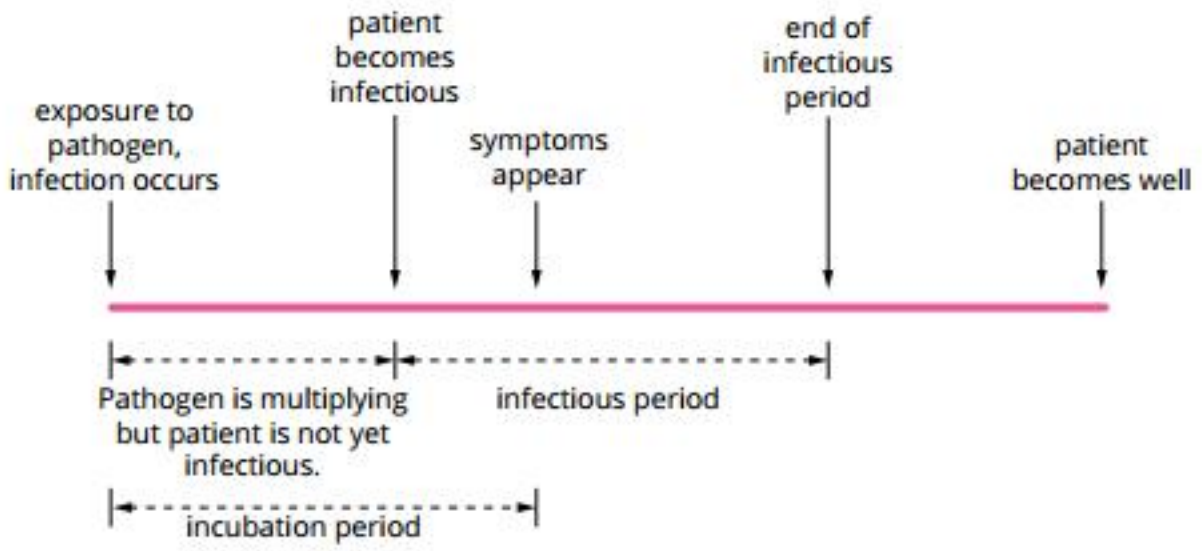


FIGURE 7.2.1 The progress of disease from exposure and infection to wellness includes several stages. The period when the pathogen is most likely to spread is between the start of infectiousness and the onset of symptoms.

The length of the incubation period can show considerable variation. This variation may be a result of the characteristics of the pathogen, such as the rate at which it reproduces, or because of traits possessed by the potential host. Some hosts resist infection more than others because of their genetics, general health, or immunity provided from vaccination or previous exposure to pathogens. Finally, the number of particles of pathogenic material that enter the host also affects the incubation period; more particles result in a shorter incubation.

At some point during the incubation period, the host becomes infectious. A long period of pre-symptomatic infectiousness helps a pathogen to infect a larger number of hosts and spread further. During the pre-symptomatic infectious period, the host feels well and continues normal activities, which brings them into contact with other potential hosts, thus spreading the pathogen more widely.

The major groups of organisms that cause disease are bacteria, protozoa (animal-like protists), oomycetes (fungus-like protists), fungi (including yeasts), several types of worms and a few arthropods.

BACTERIA

Most **bacteria** are free-living and have important ecological roles such as decomposers and as nitrogen-fixing organisms in the nitrogen cycle. Many bacteria can form dormant, temporarily non-reproductive spores in response to unfavourable conditions. With thick outer walls, the spores are resistant to many agents that destroy vegetative bacteria and may remain viable for thousands of years. If the environment becomes favourable, they recommence activity. There are bacteria that thrive under extreme conditions such as extremely high or low temperatures or pH values and high radiation or salinity, but many bacteria grow best in a moist environment with a pH close to neutral and a temperature between 20°C and 40°C.

Many bacteria are parasites of other living organisms. Some bacteria are major pathogens of crops, humans and other animals. The symptoms of bacterial disease result from the destruction of cells and tissues by bacterial enzymes, irritation by bacterial waste products, reactions to bacterial **toxins** that interfere with normal cellular functions, and the exaggerated immune responses of the sufferer to the foreign cells. Bacterial toxins can remain dangerous long after all bacteria have died. Pathogenic bacteria in humans include those that cause typhoid fever, syphilis, chlamydia, botulism, tetanus, pneumonia, diphtheria and tuberculosis. People can become carriers by being infected and carrying microbes in their body without showing any symptoms of disease.

Characteristics of bacteria

FIGURE 7.2.2 Coloured transmission electron micrograph of *Helicobacter pylori*, a flagellated spiral-shaped bacterium.

Different species of bacteria are identified by a number of physical and chemical properties such as:

- shape—cocci (spheres), bacilli (rods), spirochetes (spirals)
- organisation—single, clumps, pairs, chains
- the presence or absence of a capsule
- mobility—some have flagella or cilia to assist with movement
- requirement for oxygen—aerobic (need oxygen), obligate anaerobic (harmed by oxygen), facultative anaerobic (neither need nor are harmed by oxygen)
- nutritional requirements
- Gram staining characteristics—positive, negative.

Figures 7.2.2 and 7.2.3 illustrate some typical bacteria and their shapes and arrangements.

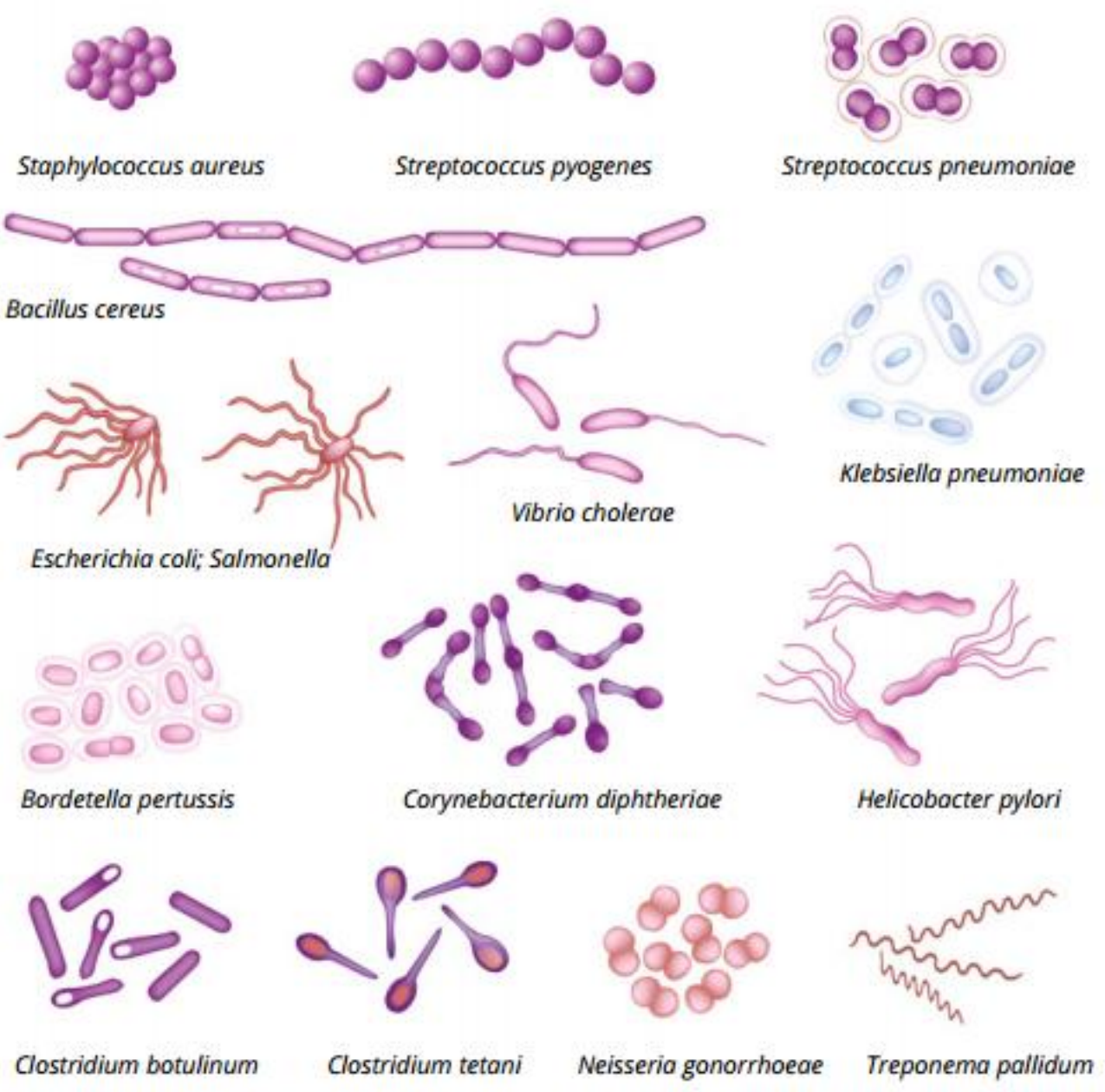


FIGURE 7.2.3 Common pathogenic bacteria, showing typical shapes and arrangements

Gram stain distinguishes between two large groups of bacteria based on differences in the chemistry and structure of their cell walls (Figure 7.2.4). It has been a key test in the initial identification of bacteria since the late 1880s and is the most widely used staining procedure. There are four steps to the Gram staining process:

1 applying a primary stain (crystal violet)
2 adding a mordant (Gram's iodine)
3 rapid decolourisation with ethanol, acetone or a mixture of both
4 counterstaining with safranin.

Gram-positive bacteria stain purple because they have a thick **peptidoglycan** (or murein) layer surrounding their cell membrane that takes up the crystal violet stain. Gram-negative bacteria stain pink because the thin peptidoglycan layer sandwiched between the inner plasma membrane and outer membrane does not take up the crystal violet as well, and the decolourisation step disrupts the outer membrane and washes the crystal violet away, enabling the pink safranin counterstain to bind to the exposed peptidoglycan layer.

Knowing whether an infection is caused by gram-positive or gram-negative bacteria informs the type of treatment used. For example, gram-negative bacteria are typically less susceptible to penicillin than gram-positive bacteria, because the outer membrane layer of gram-negative bacteria effectively excludes many substances, including certain antibiotics.

Exotoxins are highly toxic chemicals released by live bacteria into their environment. Both gram-positive and gram-negative bacteria produce exotoxins. For example, *Clostridium botulinum* shown in Figure 7.2.5a is a gram-positive, rod-shaped bacteria that produces highly lethal botulinum exotoxins, which blocks nerve function and can cause respiratory and muscular paralysis. Gram-negative bacteria also produce **endotoxins,** which are released when their cell ruptures. Endotoxins can also be fatal, but they are generally less toxic than exotoxins. The lipopolysaccharides of the gram-negative cell wall are an example of endotoxins (Figure 7.2.5b).

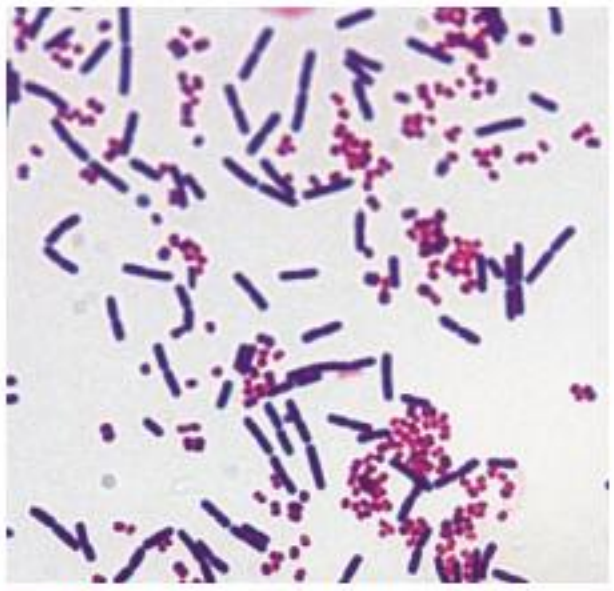


FIGURE 7.2.4 In this light micrograph, two kinds of bacteria are visible: gram-positive bacilli (rods), which have stained dark blue–purple, and gram-negative cocci (spheres), which have stained pink.

Peptidoglycan is a macromolecule made of protein and carbohydrate, found only in bacteria.

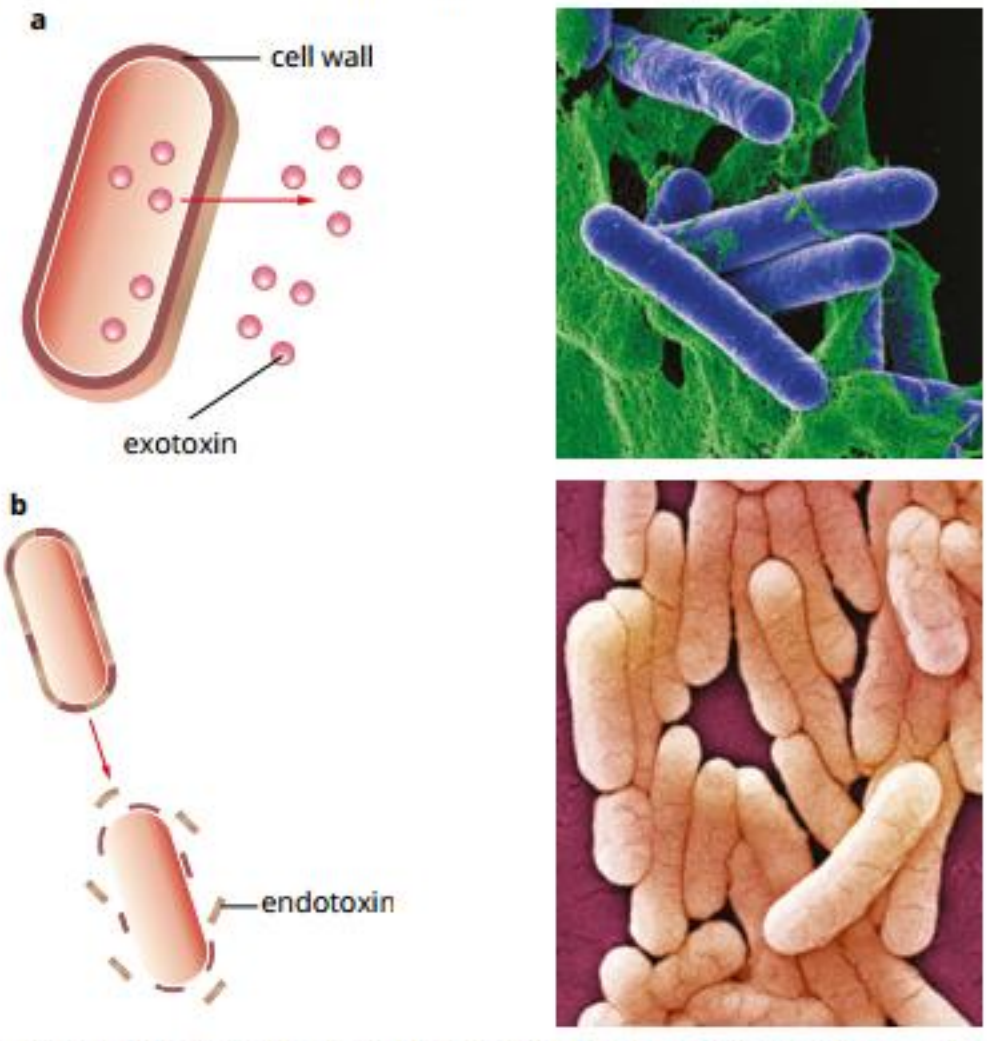


FIGURE 7.2.5 (a) Exotoxins are produced inside gram-positive bacteria as a normal part of their metabolism and are secreted into the surrounding tissues. *Clostridium botulinum* is a gram-positive bacterium that produces exotoxins. (b) Endotoxins are a part of the lipopolysaccharides of the outer membrane of gram-negative bacteria. The endotoxin is released when the bacteria die and the cell wall breaks apart. *Salmonella typhimurium* is a gram-negative bacterium that produces endotoxins.

Different toxins have different actions. Some toxins, such as the exotoxin produced by the bacterium *Clostridium tetani*, prevent the release of neurotransmitters and inhibit muscle contraction, while others such as the cholera exotoxin produced by *Vibrio cholerae* disrupt electrolyte balance in the intestines and lead to severe diarrhoea and dehydration. Both gram-positive and gram-negative bacteria can also produce exotoxins called pore-forming toxins that directly disrupt the membranes of the host's cells and cause the host cell to breakdown (lyse). By comparison, endotoxins do not disrupt host cell membranes directly, but the inflammatory response they trigger can result in indirect damage to host cells.

The lipopolysaccharides in endotoxins can produce fever in the host and increase the possibility of septic shock and circulatory collapse (extreme low blood pressure). They do this through a pathway that leads to the release of prostaglandins. The pathway to fever is shown in Figure 7.2.6. During the process, white blood cells called **macrophages** release signalling molecules called cytokines. You will learn more about macrophages and cytokines in Chapter 8. The ability of some bacteria to produce a variety of different toxins, along with antibiotic resistance, greatly increases virulence of the bacteria. Microbes can also produce enzymes that enhance their ability to invade cells and tissues. For example, the enzyme collagenase (produced by the *Aspergillus oryzae* mould and *Hathewaya histolytica* bacteria among others) breaks down collagen found in connective tissue.

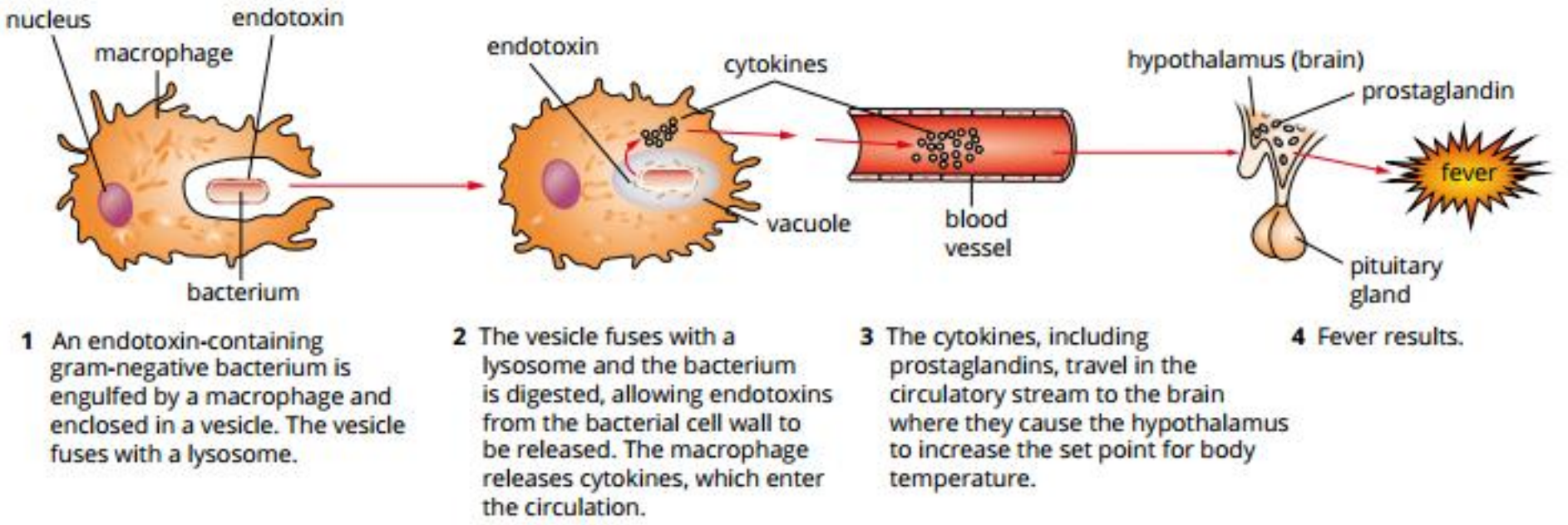

nucleus
endotoxin
macrophage
bacterium
endotoxin
cytokines
vacuole
blood vessel
hypothalamus (brain)
prostaglandin
fever
pituitary gland
1 An endotoxin-containing gram-negative bacterium is engulfed by a macrophage and enclosed in a vesicle. The vesicle fuses with a lysosome.
2 The vesicle fuses with a lysosome and the bacterium is digested, allowing endotoxins from the bacterial cell wall to be released. The macrophage releases cytokines, which enter the circulation.
3 The cytokines, including prostaglandins, travel in the circulatory stream to the brain where they cause the hypothalamus to increase the set point for body temperature.
4 Fever results.

FIGURE 7.2.6 The pathway to fever involves the release of cytokines, which signal the hypothalamus and cause the rise in body temperature.

In addition to the chemicals they produce, the virulence of microbes like bacteria depends on their structure. Bacterial invasion of the host is assisted by adhesins. These are proteins or carbohydrates on the surface of the pathogen. Adhesins help to overcome the host's mechanisms that trap and expel pathogens from the body, such as mucus or tears (see Chapter 8). Adhesins are structures that enable the bacterium to stick or adhere to host cells (Figures 7.2.7 and 7.2.8) and are one of numerous **adherence factors**.

Some adhesins are hair-like structures, called **pili** (singular: pilus), made of protein, while others are more compact structures made of carbohydrate. Gram-negative bacteria commonly rely on pili for adhesion. Gram-positive bacteria use both pili and carbohydrates. These adhesins often bind to proteins on the surface of the host cell. These host cell proteins perform a required function for the host cell, such as reception of signalling molecules like hormones or acting as protein channels. Their presence is exploited by the pathogen.

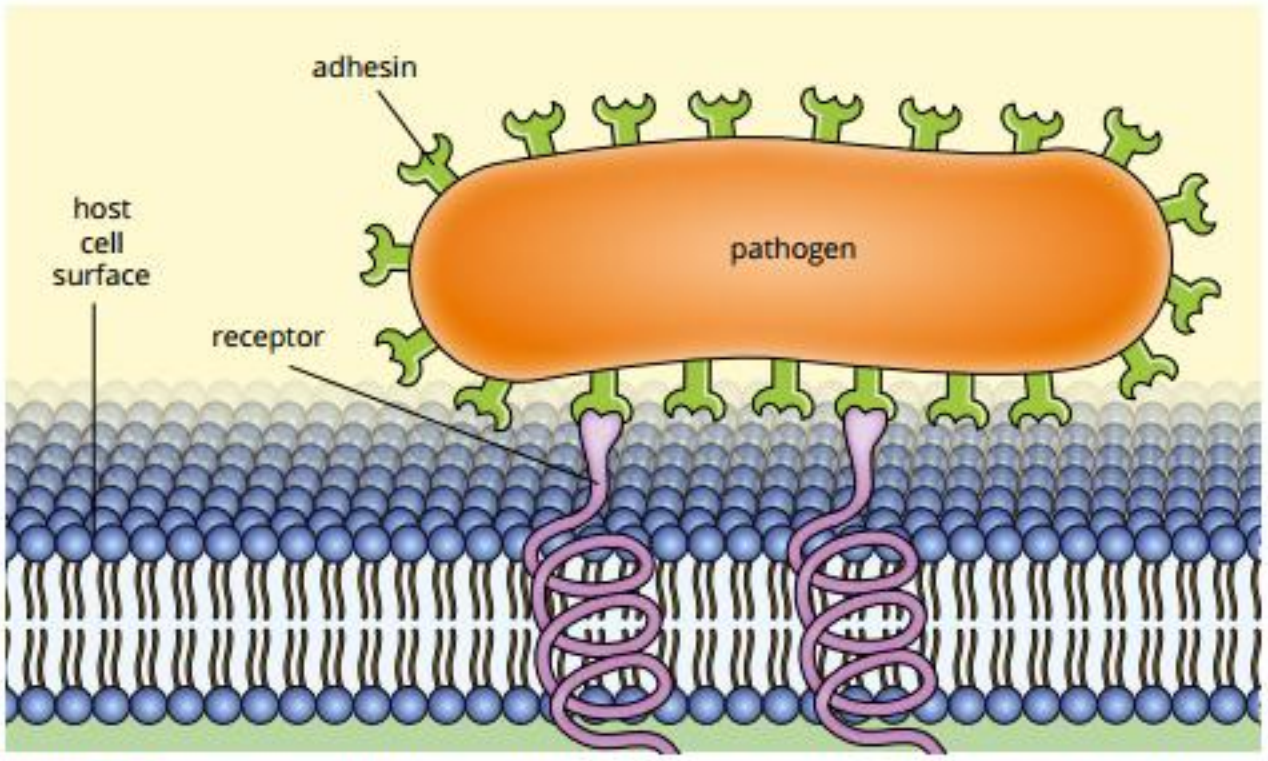


FIGURE 7.2.7 This receptor on the host cell has been used as a site for the adhesion of a pathogen.

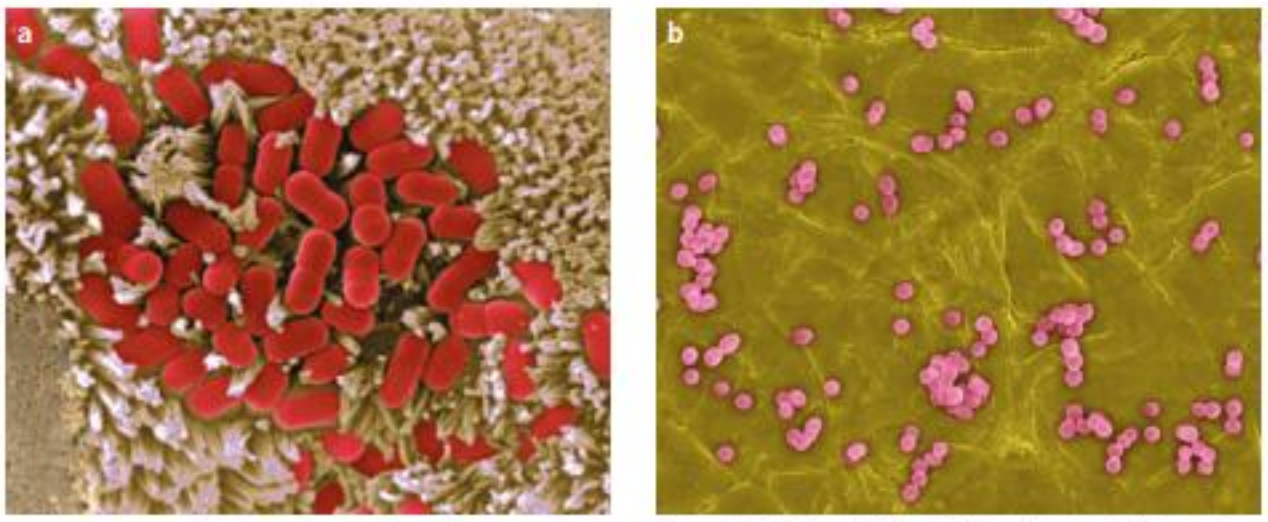


FIGURE 7.2.8 Coloured scanning electron micrographs of (a) *Escherichia coli* (bacillus, red) bacteria adhering to the small intestine, and (b) cocci bacteria (pink) adhering to skin.

Figure 7.2.9 on page 278 illustrates the structure of a bacterium. The pili and the bacterias surrounding **capsule** can be seen. Capsules are made of polysaccharides and proteins, and while not all bacteria have capsules, they contribute to the development of disease (**pathogenesis**) and virulence in multiple ways for those that do.

- Capsules provide protection from both intracellular and extracellular immune responses. For example, they block the phagocytosis of bacteria by phagocytes such as macrophages and neutrophils, and bacteria with sugary capsules, such as *Francisella tularensis*, can block the binding of complement proteins (you will learn more about these white blood cells and the complement system in Chapter 8). This evasion of the immune response allows the bacteria to persist and multiply within the host.
- Capsules help bacteria retain moisture and resist drying out (desiccation), making them more resilient in harsh environments. This enhanced ability to survive outside the host facilitates bacterial transmission and dissemination.
- Capsules can also promote adherence and the formation of protective **biofilms**, which allows bacteria to persist and resist clearance by the host immune system and antimicrobial agents. For example, the capsule of *Streptococcus pneumoniae* enhances its adherence to cells in the airways, facilitating the development of pneumonia and other respiratory tract infections. Many bacteria, such as *S. pneumoniae* and *Staphylococcus aureus*, form biofilms on surfaces within the host or on medical devices. While capsules provide structural integrity that promotes biofilm formation, they are not necessary for biofilms to form.

Biofilms are structured communities of microbes embedded in a self-produced extracellular matrix of polysaccharides, proteins and DNA. While bacteria are the most common components of biofilms, other microorganisms such as fungi, algae and protozoa can also contribute to or form biofilms independently.

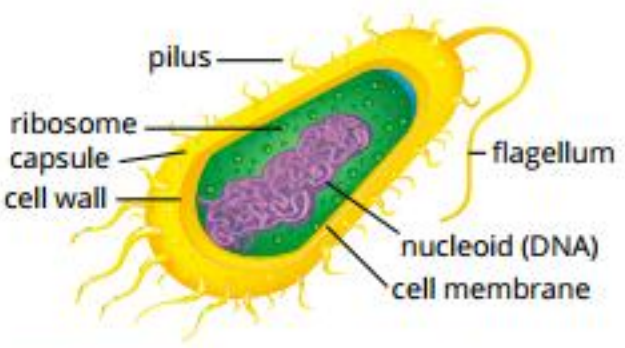


FIGURE 7.2.9 Many bacteria have pili, which assist in adhesion, and a capsule, which helps them evade the immune system of host organisms.

Understanding how capsules mediate adherence can inform the design of antimicrobial coatings for medical devices to prevent bacterial colonisation and biofilm formation, and antimicrobial agents can be used to disrupt capsule synthesis or function. Vaccines can also be developed that induce immune responses against capsule antigens (e.g. polysaccharide conjugate vaccines for *Streptococcus pneumoniae* and *Haemophilus influenzae*). The targeting of bacterial capsules through interventions such as these can reduce the burden of bacterial diseases.

Antibiotic resistance is also becoming a critical problem as bacteria with antibiotic resistance are being selected through antibiotic-rich environments. In any population of bacteria, some individuals will have a natural resistance to antibiotics. In an antibiotic-rich environment, these individuals are the ones most likely to survive and reproduce. This creates highly resistant strains. The problem is exacerbated by the misuse and over-use of antibiotic medications because this allows the survival of a few highly resistant bacteria, which then increase in numbers. Bacteria can also transfer genes, including those that confer antibiotic resistance, to other strains of previously non-resistant bacteria.

Plants and pathogenic bacteria

Infection by bacteria is not confined to animals. Plants can also be infected by pathogenic bacteria.

The bacterium *Agrobacterium tumefaciens* infects plant roots, where it causes cells to proliferate. Figure 7.2.10 illustrates the cycle of infection and disease in infected plants. Crown gall bacteria enter the plant through a fresh wound and multiply in intercellular spaces. They produce large amounts of a plant growth hormone called auxin, which causes nearby plant cells to multiply and produce nutrients that only the bacteria can use. These rapidly dividing plant cells form a **gall** (an abnormal growth, similar to a tumour), which continues to grow out of control (similar to cancer cells), producing food for the pathogen but preventing normal growth of the plant. As the gall develops, the plant's vascular tissues (xylem and phloem) become blocked, which compromises its ability to move water and nutrients, and results in reduced growth, death of parts of the plant or, in cases of major infection, the death of the whole plant.

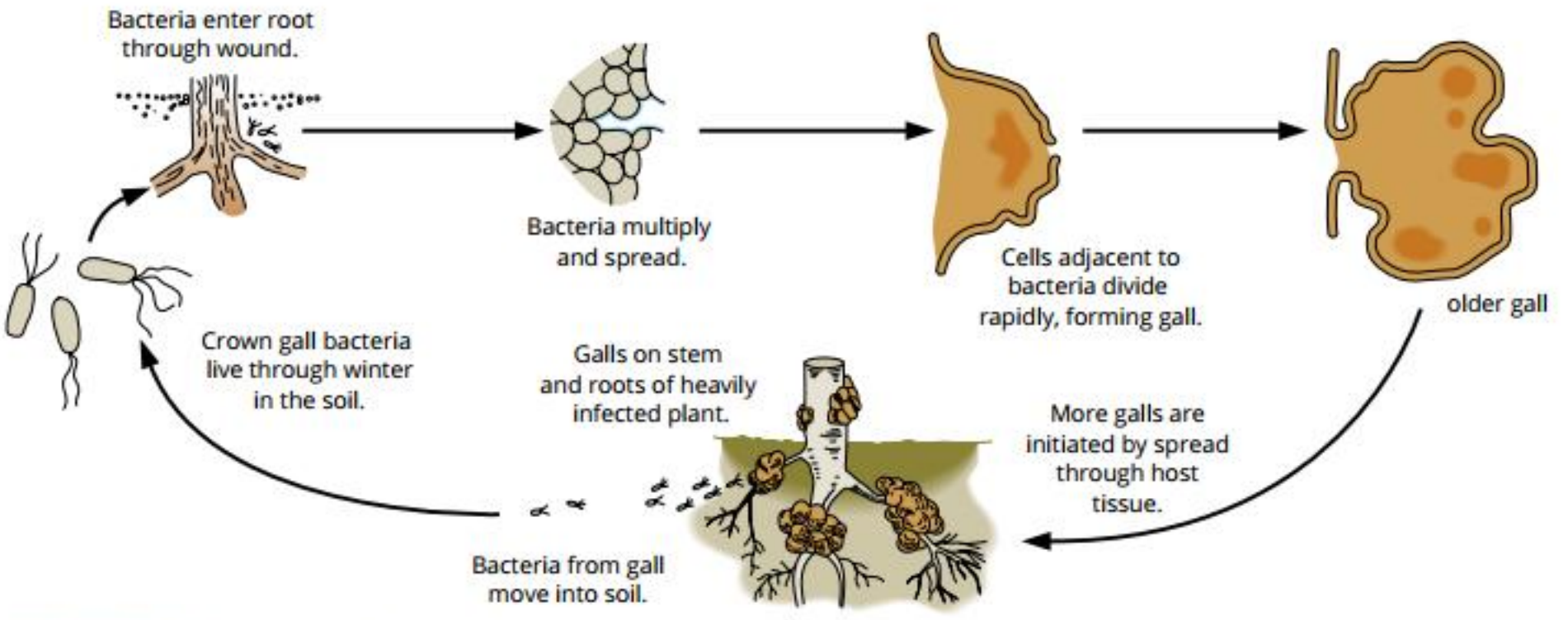

Bacteria enter root through wound.
Bacteria multiply and spread.
Cells adjacent to bacteria divide rapidly, forming gall.
older gall
More galls are initiated by spread through host tissue.
Galls on stem and roots of heavily infected plant.
Bacteria from gall move into soil.
Crown gall bacteria live through winter in the soil.

FIGURE 7.2.10 The disease cycle of Crown gall, which is caused by *Agrobacterium tumefaciens*.

CASE STUDY 7.2.1

Legionnaire's disease—tracking the cause

Legionnaire's disease is caused by bacteria in the genus *Legionella*. As recently as 1976, this disease was completely unknown. The first recognised case of Legionnaire's disease occurred after a convention held in Philadelphia, USA, in 1976. After the convention, 221 people became ill and 34 of them died. They presented with a pneumonia-like illness that also included diarrhoea, nausea and confusion. As reports of deaths from an unknown pneumonia-type illness came into the Centers for Disease Control (CDC), research began to identify the cause. The first commonality identified between the affected people, even though they were now spread throughout the USA, was that they had all either attended the Philadelphia convention or had been working or visiting in the area. This allowed the scientists to narrow the outbreak down to the hotel in Philadelphia where the convention was held.

To further narrow down the cause, the disease detectives interviewed the surviving victims and their families, along with a number of people who had also been at the convention but who had no sign of illness.

Each method of disease spread was considered:

- Contaminated food—along with the food served in the hotel, there were many restaurants and cafes in the area, most of which had been visited by at least some of the patients and the healthy controls, but there was no pattern that pointed to a particular food source.
- Waterborne—both patients and controls were asked about drinking un-bottled water or consuming ice. There was no link to ice consumption, but a higher rate of water consumption in the ill than in the healthy was observed. The difference was statistically significant, but drinking water could not be used to account for all the cases.
- Direct person-to-person contact—this was ruled out because there was no clustering of cases among families or people who had shared rooms at the convention.
- Vectors—no evidence was found to suggest that any of the patients had come in contact with animals, and none complained of suffering from insect bites.
- Airborne—having ruled out all other sources, in at least some cases, the scientists concluded the most likely method of transmission was airborne with some contamination of drinking water from the air. They determined that the cause was most likely inhalation into the lungs, causing the pneumonia symptoms. This conclusion was further supported by the fact that cigarette smokers were over-represented in the group with the disease. Lung damage caused by smoking has been shown to increase susceptibility to lung infection by bacteria. The data in Table 7.2.1 shows that the mean increase in the probability of a smoker developing Legionnaire's disease, if exposed, is 3.6 compared to non-smokers.

TABLE 7.2.1 Bacterial infections of the respiratory tract associated with smoking. Odds ratio is a measure of the association between an exposure and an outcome.

Infection	Odds ratio (95% confidence interval)
Nasopharyngeal and respiratory pathogens (e.g. *Streptococcus pneumoniae, Haemophilus influenzae, Legionella pneumophila*)	2.5 (1.1–6.0)
Legionnaire's disease	3.6 (2.1–5.8)
pneumonia	2.6 (1.9–3.5)

Having decided that an airborne pathogen of unknown type was likely responsible for the outbreak, researchers continued to try to find the cause of disease. The researchers tested recovering patients' blood for the presence of the same **antibody**. Antibodies are proteins made during the immune response to a particular pathogen. You will learn more about antibodies in Chapter 8.

Antibodies in the recovering patients were compared with antibody samples taken from the hotel employees. The presence of the same antibodies in the employees who had not been ill during the outbreak suggested that this disease had been prevalent in the area for some time before the convention and that exposure did not necessarily cause a severe illness.

It took several months to identify the cause of the disease as *Legionella* bacteria. By then, the source at the convention hotel could not be identified, but it was probably the air-conditioning towers. Individuals who stood near vents would have been exposed to more airborne bacteria than those who stood further away, and some individuals were also more susceptible because of other health issues, age, or smoking.

After identifying the cause of the outbreak, scientists searched records and other outbreaks of similar character were identified and are now ascribed to Legionnaire's disease. *Legionella* outbreaks continue to occur, usually through improperly decontaminated air-conditioning towers.

Review

1 Describe the methods used to track down the cause of Legionnaire's disease.

2 Explain why poorly maintained air-conditioning towers can lead to outbreaks of bacterial diseases.

PROTISTS

Protists are a remarkably diverse group of organisms. They range from multicellular photosynthetic organisms, such as seaweed, to highly pathogenic, unicellular organisms, such as the **plasmodium** that causes malaria. Three groups of protists have members that are pathogenic to animals:

- zooflagellates (e.g. *Trypanosoma*, which causes sleeping sickness, *Giardia*, a common cause of diarrhoea, and *Leishmania* (Figure 7.2.11a), which causes leishmaniasis)
- sporozoans (e.g. *Plasmodium* (Figure 7.2.11b), which causes malaria)
- sarcodines (e.g. *Entamoeba histolytica* (Figure 7.2.11c), which causes amoebic dysentery).

Another group of protists—oomycetes—are primarily pathogenic to plants.

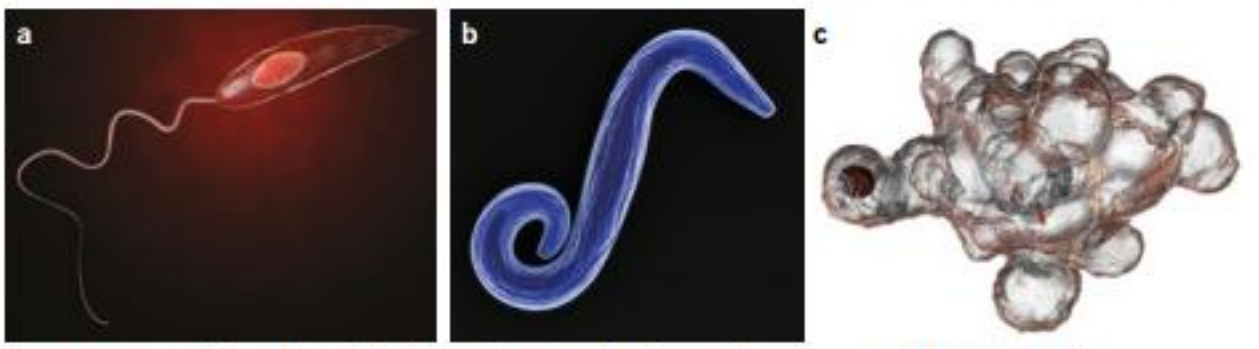


FIGURE 7.2.11 Examples of the three classes of pathogenic protozoans: (a) *Leishmania*, (b) *Plasmodium* and (c) *Entamoeba*.

Pathogenic sporozoan: malaria

Malaria is a disease that occurs across large areas of the world. It causes up to 1.8 million deaths a year, with more than half being of children aged under five years. Figures from different sources vary significantly because many factors are involved, so it is difficult to determine whether a death is due to malaria or another cause. In many cases, malnutrition, other diseases and malaria combine to cause death.

There are several species of plasmodium that cause malaria. The most common are *Plasmodium falciparum* and *P. vivax*. *P. falciparum* is linked to higher levels of mortality than *P. vivax*. Figure 7.2.12 shows the distribution of these two species.

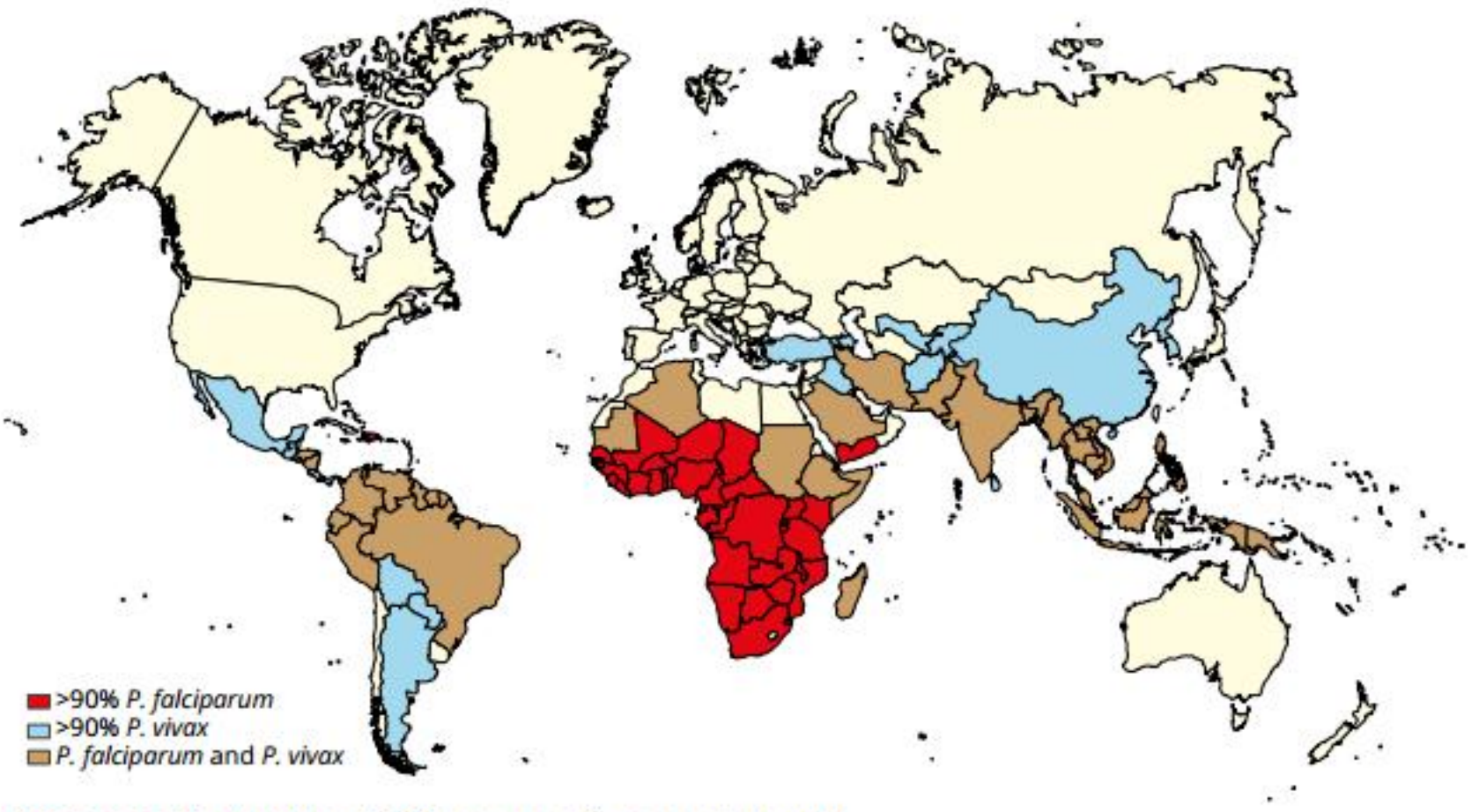


FIGURE 7.2.12 The distribution of *P. falciparum* and *P. vivax* across the world.

There are two hosts in the life cycle of *Plasmodium* parasites, as can be observed in Figure 7.2.13, which illustrates the various stages of the malarial life cycle. The adult stage occurs in humans (the primary host). After a person is bitten by an infected female mosquito (only female mosquitoes feed on blood), *Plasmodium* larvae pass in the blood to the liver, where they feed and multiply asexually for two weeks. The larvae then leave the liver and pass into the bloodstream and infect red blood cells. Inside the red blood cell, the parasite grows and feeds, almost filling the cell, before the parasite splits into many tiny larvae. It is then that symptoms associated with malarial infection begin to appear. Symptoms include recurring bouts of fever every few days, with high temperature, sweating, shivering and delirium.

Malarial attacks are associated with the bursting of red blood cells that spill the parasites, which at this stage of their life cycle are called **merozoites**, into the circulatory system. The presence of one million merozoites in the blood is sufficient to produce symptoms of malaria. As the red blood cells rupture, the oxygen-carrying capacity of the blood is compromised, causing fatigue. Rupture of cells in any part of the body initiates the symptoms of inflammation, which include fever and chills. You will learn more about this process in Chapter 8, Immune Response.

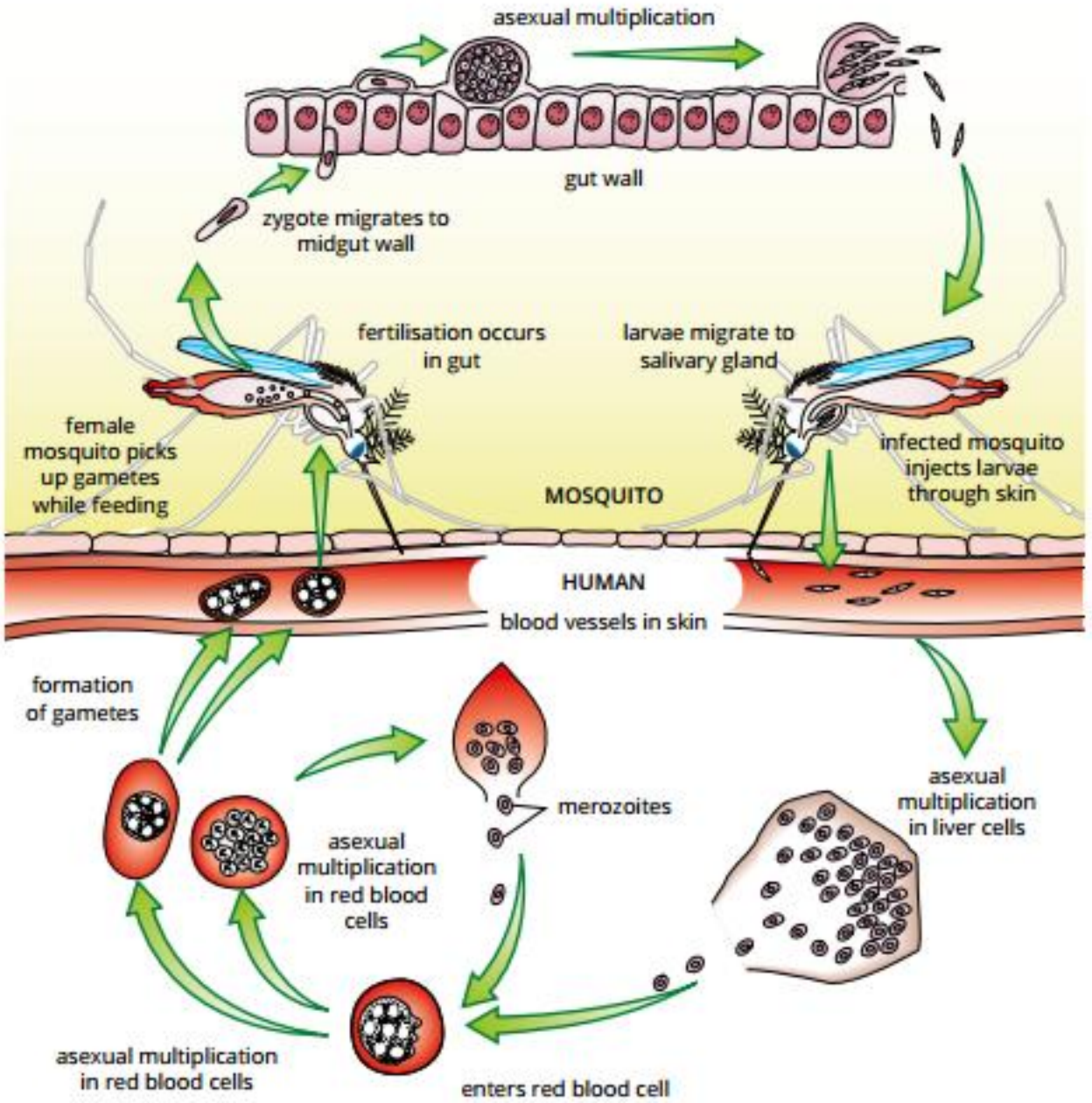


FIGURE 7.2.13 The life cycle of the malarial parasite (*Plasmodium* sp.) involves two hosts, with humans being the primary host.

Fevers occur in regular cycles because merozoites take a fixed period (1, 2 or 3 days, according to the species of parasite) to grow, mature and divide asexually inside red blood cells, which then all burst together. The newly released merozoites attach to and infect more red blood cells. They grow for the same period within the red blood cells, then burst out, and the cycle continues. While it has not been shown definitively, there is evidence that the malarial merozoite uses a protein found on the surface of red blood cells, called **Duffy**, to enable it to attach to the cells and push its way inside. The merozoite does this by a process similar to endocytosis. This enables the parasite inside the red blood cell to be enclosed in a vesicle. Unlike endocytosis, this process is not controlled by the red blood cell; rather, the merozoite is in charge.

During the process of merozoite invasion of red blood cells, some of the merozoites in the bloodstream develop into a gamete, producing forms that can be ingested by mosquitoes and transferred to a new host. The development of the gamete-producing stage tends to follow a circadian rhythm. They are found in the blood at times of the day when the intermediate host, the *Anopheles* mosquito, is feeding, which is usually in the early evening and night. When taken up with blood, sexual reproduction occurs in the midgut of the female mosquito when gametes fertilise and develop into larvae, which migrate through the stomach wall and into the salivary glands of the mosquito, where the larvae then undergo asexual reproduction. In the salivary gland, a single larva can multiply asexually to produce 10 000 larvae, which are passed on to a primary host during feeding.

Malaria can only be transmitted in regions where the *Anopheles* mosquito lives. There are several species of *Anopheles*. Most of the species can transmit malaria, but how likely they are to infect humans varies enormously. Some species prefer the blood of other animals over humans, and vice versa. Two species of *Anopheles* in Africa, *A. gambiae* and *A. funestus,* show a distinct preference for human blood, which is probably one factor leading to the high prevalence of malaria in Africa.

Mosquitoes rely on standing water for their eggs to hatch and their larvae to mature. Therefore, rainfall is a significant factor in malarial spread (Figure 7.2.14). Once the mosquito larvae have matured into adults, they must bite an already infected person and ingest the gametes of the malarial parasite. The gametes form a zygote.

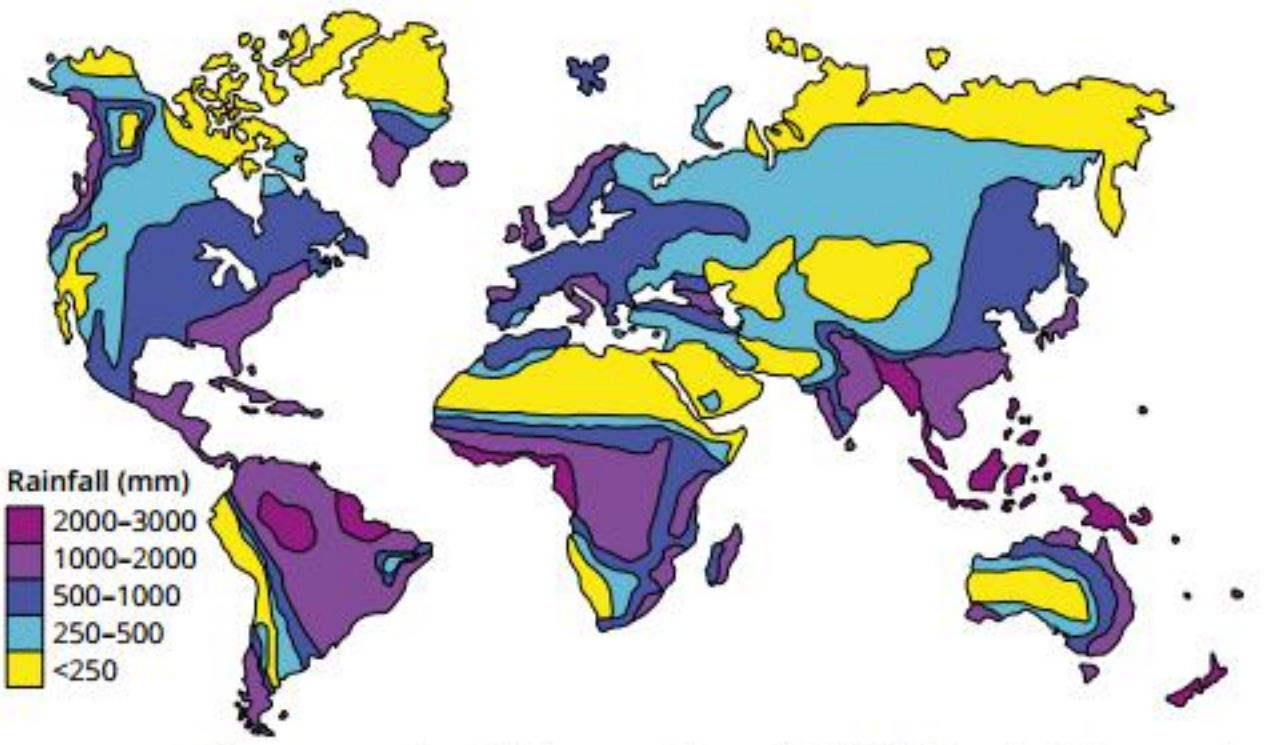


FIGURE 7.2.14 The mean annual precipitation across the world. Rainfall is important in the spread of malaria.

The plasmodium then takes 9–21 days to complete its life cycle, ready to produce gametes, infect the next mosquito and subsequently the next human host. The length of time depends on the ambient temperature. In cooler areas the process takes longer. Studies of the two species have shown that this part of the mosquito's life cycle cannot occur at temperatures below 15°C for *P. vivax* or below 20°C for *P. falciparum*. The ideal temperature for this stage is above 25°C. Consequently, in areas where temperatures frequently fall below 15°C, transmission of malaria is highly unlikely. Figure 7.2.15 shows the mean temperatures in January (Figure 7.2.15a) and in July (Figure 7.2.15b). Temperature and rates of transmission are strongly correlated.

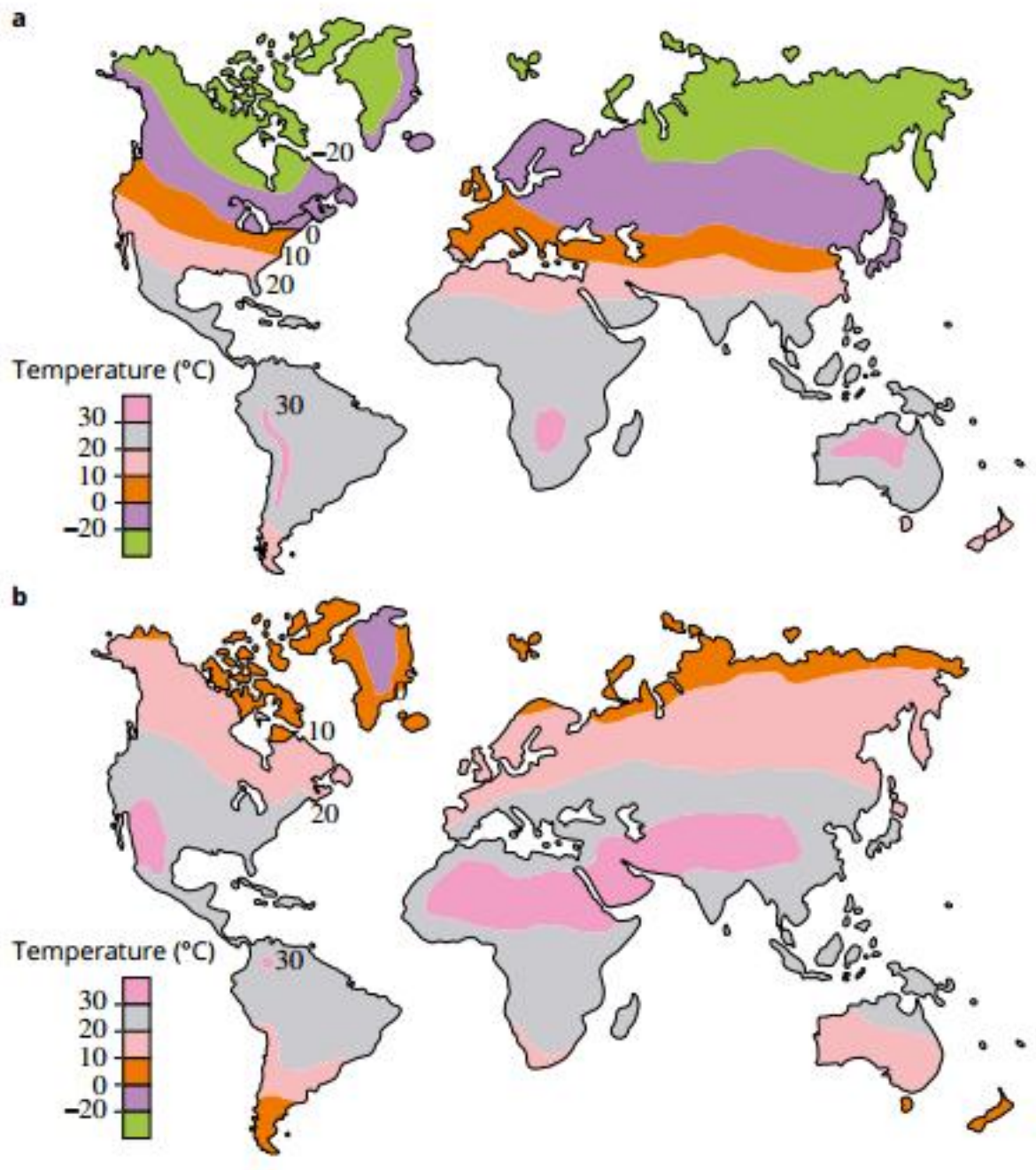


FIGURE 7.2.15 Mean annual temperatures worldwide in (a) January and (b) July. Temperature is important in the development and spread of malaria.

Pathogenic oomycetes

The **oomycetes** (phylum Oomycota) include organisms that cause blight and downy mildew on plants. Long thought of as fungi, the oomycetes are now classified in the kingdom Protista. They have motile cells (with flagella), walls of cellulose, and many cellular processes that are not found in fungi. When spores are released on a leaf, they may be carried in water droplets to other leaves, swim to a germination site, or germinate directly, sending out **hyphae** (thread-like structures) that branch and invade plant tissue.

FIGURE 7.2.16 This eucalypt forest in the Brisbane Ranges National Park in Victoria is infected with *Phytophthora cinnamomi*. Many species of native Australian plants such as the grass trees shown are highly susceptible to this pathogen and rapidly turn brown and die.

These branching hyphae (also called haustoria) penetrate living cells and absorb nutrients or release enzymes that digest cytoplasm into molecules that can be absorbed. In damp weather, some hyphae grow out through stomata and form spores at their tips. When ripe, spores are blown by the wind to re-infect the same host and to infect other plants.

Members of the oomycetes are important plant pathogens. The genus *Phytophthora* (which means 'plant destroyer') is of particular concern because it has destroyed many crop species and native plants (Figure 7.2.16). There are about 35 species of *Phytophthora*, which infect many crops, including potato, tomato, apple, tobacco plants and citrus trees. In Australia, *Phytophthora cinnamomi* has destroyed tens of thousands of hectares of valuable eucalypt timberland. The spores of *P. cinnamomi* can survive for years in moist soil and are attracted to the roots of the plants they infect by a chemical released from the roots.

P. infestans caused the 1845–49 potato blight in Ireland. The disease caused the potato crop to fail, resulting in a famine during which more than a million people died of starvation or diseases that thrived in the malnourished population. Up to two million people emigrated from Ireland to escape the famine; many came to Australia.

Phytophthora has a complex life cycle, with a sexual and an asexual (Figure 7.2.17) stage. The spores may be transmitted from plant to plant in water and soil, by wind or vectors. It thrives under wet conditions, especially where soil is poorly drained. It is easily transported in wet soil attached to boots, tyres, or machinery. The motile **zoospores** can swim through soil water to infect new plants. The spores enter the plant through any damaged area of the roots, and produce hyphae that grow through the plant, blocking the xylem and phloem, reducing the movement of water and nutrients around the plant, and ultimately killing the plant in susceptible species. Figure 7.2.18 shows the specialised hypha (sporangiophore) of *P. infestans* growing out of the stomata of a leaf. At some ends are bulb-shaped structures (sporangia), which contain and release **chlamydospores**. These spores can be blown by the wind or transported by other organisms for long distances. When they fall in a suitably moist environment, they mature and release motile zoospores, which continue the cycle of infection.

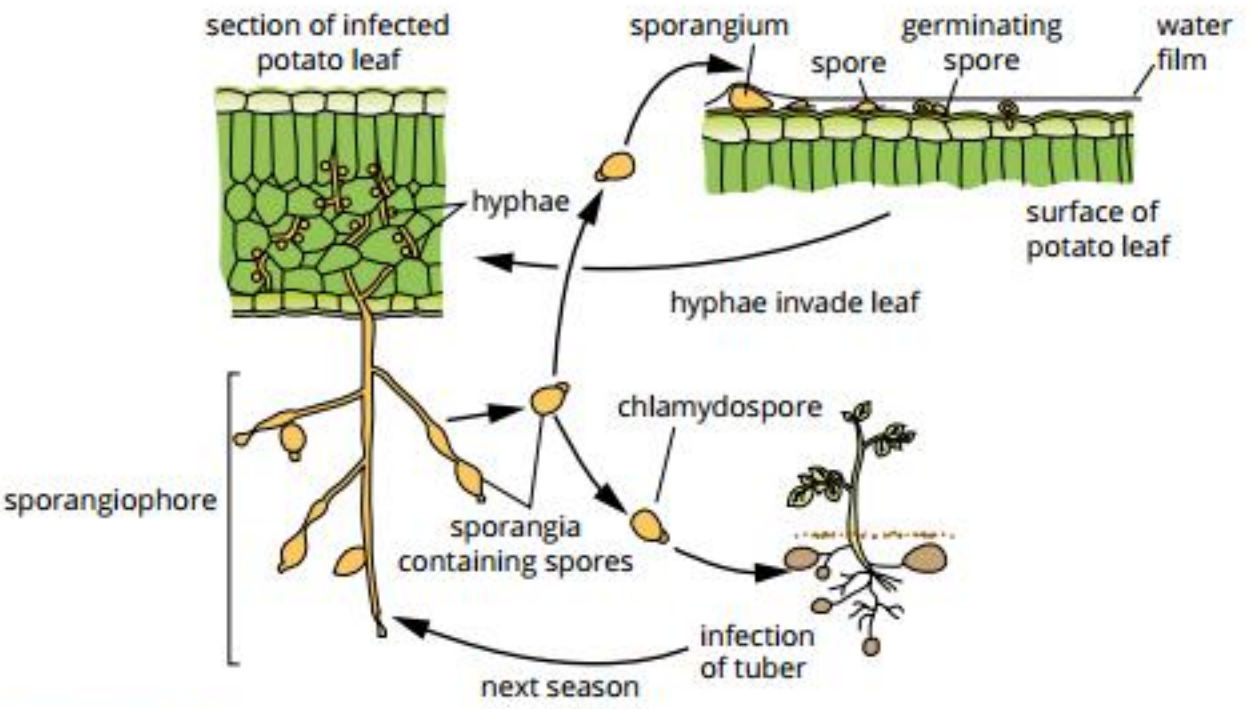


FIGURE 7.2.17 The asexual disease cycle of *Phytophthora*. Spores invade roots and leaves and germinate, and then hyphae grow out of stomata and release more spores to infect other plants.

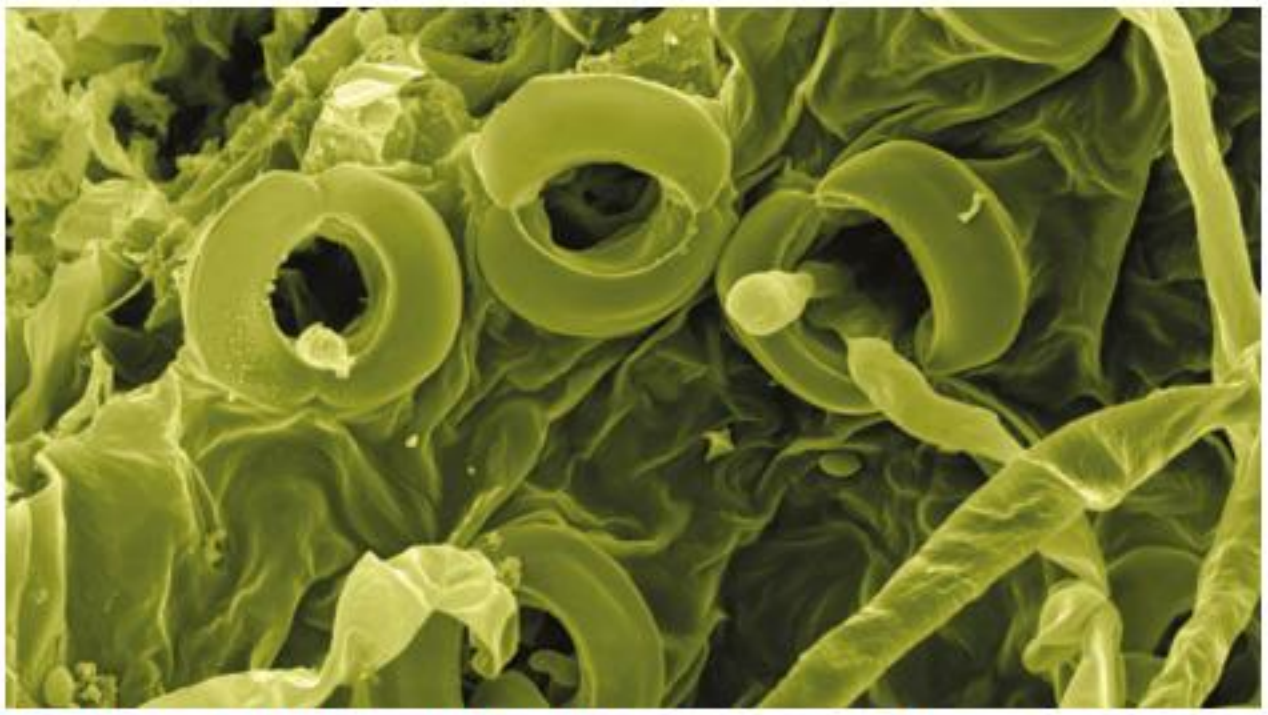


FIGURE 7.2.18 A coloured scanning electron micrograph of *Phytophthora infestans* hyphae with spore cases attached growing through the stomata of an infected leaf.

FIGURE 7.2.19 A skin infection caused by *Pythium insidiosum*. In this case, the infection is in a horse, but humans and other mammals can also be infected.

Only one member of the oomycetes, *Pythium insidiosum*, is known to be pathogenic to mammals, including humans. Infection in humans is rare but when it does occur it causes a serious disease, pythiosis, which is shown in Figure 7.2.19.

FUNGI

Fungi are a diverse kingdom of organisms, ranging from the macroscopic to the microscopic (moulds, unicellular yeasts and yeast-like fungi). Fungi can secrete digestive enzymes and other chemicals into their environment to break down organic matter, which can then be absorbed into the **fungus**. It is these secreted substances that are usually responsible for causing disease in animals and plants. Some fungi, such as the *Amanita muscaria* illustrated in Figure 7.2.20, produce biologically active chemicals called alkaloids, some of which cause disease symptoms due to poisoning if ingested. Fungal cells produce surface glycoproteins and polysaccharides that act as **antigens** (substances on the surface of cells that can trigger an immune response), allowing them to be identified by cells of the immune system.

Fungal pathogens

Many fungi are plant pathogens, causing diseases such as leaf spot in eucalypts, rust and ergot in cereals, and Dutch elm disease. In humans, fungi can cause athlete's foot (tinea), ringworm and thrush. Certain species of fungi are insect-killers. For example, when fungal spores of *Ophiocordyceps unilateralis* land on an ant, they grow through its skin and branch through the insect, eventually killing it (Figure 7.2.21). The hyphae then form fruiting bodies that shoot masses of spores away from the insect.

FIGURE 7.2.20 Many field fungi, such as *Amanita muscaria*, contain toxic alkaloids.

FIGURE 7.2.21 *Ophiocordyceps unilateralis* is a fungus that infects ants.

Only a few fungi are pathogenic to animals. Three main groups cause disease in humans:

- moulds, which are filamentous
- true yeasts, which are unicellular
- fungi-like yeasts, which are like yeasts but may form long non-branching filaments.

The most well-known, but not particularly harmful, are the fungi that infect the skin. Different forms of the ringworm fungus cause tinea, which grows in the moist warmth between the toes, and ringworm of the scalp, body and toenails. Thrush, endocarditis and septicaemia are caused by the fungus-like yeast *Candida albicans* (Figure 7.2.22). Direct contact is usually involved in transmission of these skin pathogens. People experiencing acquired immunodeficiency syndrome (AIDS) resulting from untreated human immunodeficiency virus (HIV), or other diseases in which their immune responses are impaired, often suffer from fungal infections.

FIGURE 7.2.22 A coloured scanning electron micrograph of the yeast *C. albicans*, which causes thrush (candidiasis). Depending on environmental conditions, *C. albicans* takes a unicellular yeast-like form or a multicellular filamentous form.

CASE STUDY 7.2.2

Panama disease

Many crop species are susceptible to fungal infections. In 2016, the Queensland banana crop was under threat from Panama disease. This is caused by a strain of the fungus *Fusarium oxysporum* f.sp. *cubense*. There are many strains of *F. oxysporum*. Some are more pathogenic than others. The different forms are denoted f.sp. (or *formae speciale*).

The *cubense* form of fungus first appeared in Cuba, hence the name. Within *Fusarium oxysporum* f.sp. *cubense* are a number of subgroups or races. The race that infects bananas is Race 4, which has a particular variant that only seems to appear in the tropics, hence Tropical Race 4.

The most popular banana in production was the Gros Michel until the early 1960s when the world crop was devastated by an as yet unidentified strain of *Fusarium*. A search for resistant banana strains identified the now popular Cavendish bananas.

In 1989, a new strain of *Fusarium oxysporum* appeared in bananas in Malaysia and Indonesia. This strain, VGC 01213 or Tropical Race 4 (TR4), is highly pathogenic to Cavendish and many other banana varieties. The spread of Fusarium TR4 has been documented and a timeline is shown in Figure 7.2.23. Although identified in 1989 in Malaysia and Indonesia, it is thought that *F. oxysporum* may have been present for some time before then.

FIGURE 7.2.23 A timeline of the spread of *Fusarium oxysporum* Tropical Race 4 (TR4)

Panama TR4 has the potential to wipe out banana production worldwide. The fungus initially enters the plant where roots have been damaged by borers, worms, or mechanical damage, or through direct contact of the roots with the roots of other nearby infected plants. Young plants are particularly susceptible to infection. Once in the root, the fungus produces many spores, which are drawn up the plant in the xylem. Along the xylem are pits that allow water to leave the xylem and move into the tissues. These pits become blocked by the spores, which then geminate and start to grow hyphae into adjacent xylem vessels. The hyphae then produce more spores. The whole of the vascular system becomes impregnated with the fungus, blocking water flow and ultimately causing the death of the plant. Banana plants in Figure 7.2.24 show signs of the leaf dieback typical of *Fusarium* infection. As the plant dies, the hyphae grow out of the xylem and into the dying and dead tissue, where chlamydospores are produced. These spores end up in the soil where they can lie dormant for many years. Soil testing has shown that fields that have not had banana plants in them for 20 years still contain viable chlamydospores.

FIGURE 7.2.24 These banana plants are showing the leaf dieback that is symptomatic of *Fusarium* infection.

Review

1 Describe how *Fusarium* causes the death of the banana plants that it infects.

2 **a** Propose possible strategies that should be undertaken to limit the chances of *Fusarium* spreading from currently infected plantations.

b Argue the likelihood of the proposed strategies for success over the long term (e.g. decades).

CASE STUDY 7.2.3

Black stem rust—a wheat pathogen

Another serious crop-infecting fungus is *Puccinia graminis* (black stem rust), which infects wheat. In the northern hemisphere, this fungus has a complex life cycle involving two hosts and several different types of spores. Thick-walled black spores on wheat germinate after winter and produce new spores (**basidiospores**) in spring. If basidiospores fall on the alternative host (barberry), they germinate, invade the host tissue, and produce haploid fungal spores that fuse and produce a fruiting body. Different spores (**aeciospores**) are then released and dispersed by the wind. If the aeciospores land on a wheat plant, they germinate and invade the wheat plant by entering through stomata on stems or leaves, causing severe damage to the plant. Inside the wheat plant, the aeciospores grow into another type of fruiting body, which produces many spores that reinfect other wheat plants. Late in summer, fruiting bodies form and produce resting spores that become basidiospores the following spring. In Australia, the cycle is limited to the asexual re-infection of wheat and related grasses. The left side of Figure 7.2.25 shows the part of the life cycle of *P. graminis* completed within Australia. The lack of the necessary host plant, barberry, means that the sexual part of its life cycle is unable to occur.

Plant breeders are continually developing new varieties of wheat that are resistant to fungal diseases. This is difficult because the fungi causing these diseases are continually changing, evolving into new strains that can infect the new varieties.

Review

1 **a** State the name given to the process of fusion of the haploid fungal cells.
b Analyse Figure 7.2.25 and identify the type of spores that are formed as a result of sexual reproduction.

2 Explain how the lack of the sexual part of its life cycle will affect the fungus and propose how this could be of advantage to Australian farmers.

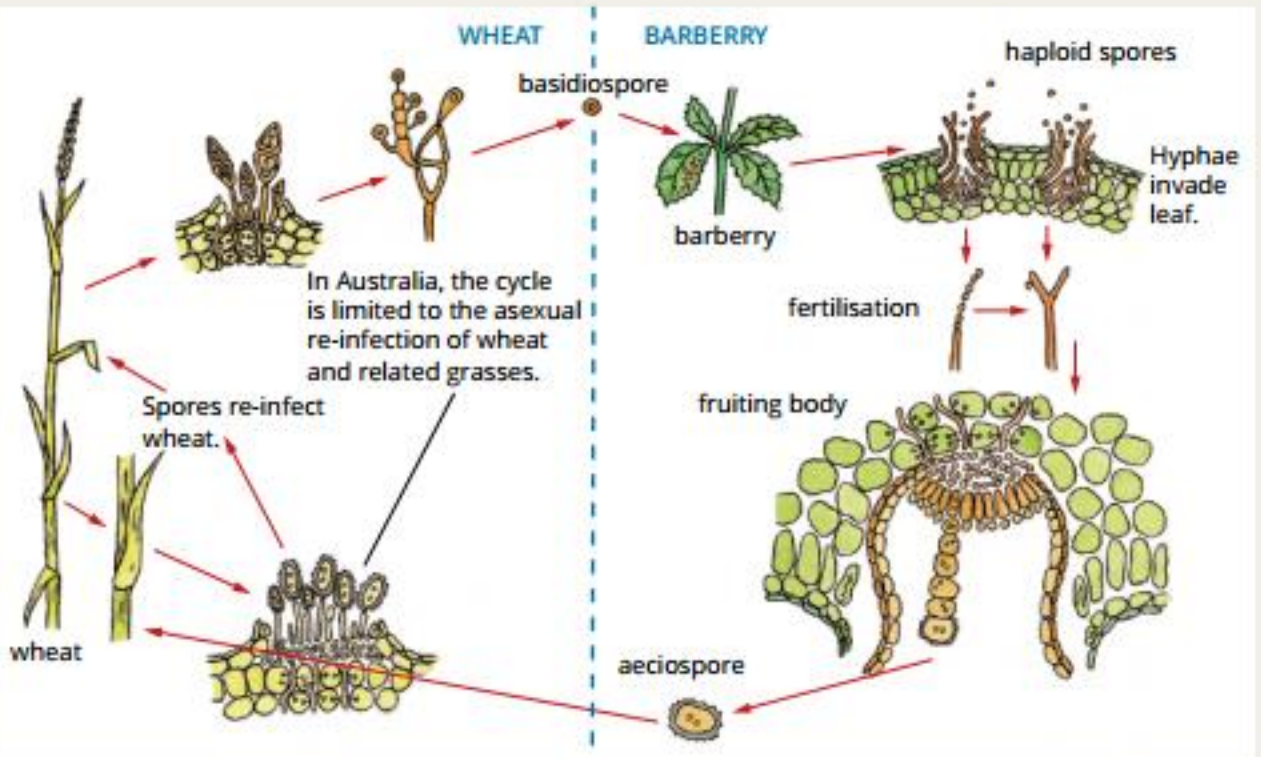


FIGURE 7.2.25 The complete life cycle of *Puccinia graminis* involves two hosts. In Australia, only the asexual part of the life cycle, which occurs in wheat, occurs because of the warmer climate and the lack of the barberry host.

Ergotism

Some fungi produce toxins that can be extremely poisonous to humans and animals. The earliest reported toxic fungus was *Claviceps purpurea*, a parasite that infects the kernel of cereal grains, particularly rye, and produces the substance ergot. It is best known, not for its damage to the rye crop, which is relatively mild, but for its effects on mammals when eaten. Human poisoning (**ergotism**) due to consumption of infected rye grain dates back to before the Middle Ages. There are two types of ergotism. Gangrenous ergotism affects circulation to the extremities, causing gangrene (and some gastrointestinal problems). Convulsive ergotism affects the skeleto-muscular and nervous systems, causing convulsions and brain and spinal lesions.

One of the chemicals in ergot is lysergic acid. This chemical has much in common with the hallucinogen LSD (lysergic acid diethylamide) and many of the psychiatric effects of ergot can be attributed to lysergic acid. The chemicals produced by ergot also affect blood vessels. They stimulate muscles in arterioles to contract, thereby reducing the diameter of the blood vessel. This reduces blood supply and starves the tissues of oxygen, resulting in cell death. Feet and hands are usually affected first. Digestive tracts may also show symptoms of reduced blood flow, but this is less common. Loss of blood flow to skeletal muscles may result in convulsions caused by the muscles cramping.

Like most poisoning diseases, ergot poisoning is dose dependent. Higher doses result in more serious symptoms. Ergot contains ergotamine, a chemical that is chemically similar to the neurotransmitters serotonin, dopamine and noradrenaline. Its similarity to these important signalling molecules explains its action, because it can take the place of a neurotransmitter and stimulate a response in muscles and organs.

Ergot poisoning is rare in humans today but can cause serious problems for farmers if it infects feed. Its effects on blood flow in the uterus cause females to miscarry. Reduction in blood flow can result in decreased milk production in dairy cattle. Also, sheep and cattle can develop gangrene of the feet, leading to their death.

On the beneficial side, in very small doses ergot has proved useful in the treatment of migraine headaches, and for inducing birth and reducing post-birth bleeding, as it causes both uterine blood vessels and the smooth muscles of the uterus itself to contract.

Most pathogenic fungi do not cause serious illnesses in normally healthy humans, but they can cause serious infections in individuals with compromised immune systems, such as people experiencing AIDS from untreated HIV, taking drugs that suppress their immune system, or who are seriously ill with other infections. At times, growth of *Aspergillus* fungi has caused issues in hospitals. In April 2015, an *Aspergillus* infestation forced the closure of the intensive care ward of in Queensland?

OTHER PARASITES

Other parasites also cause disease in humans. Parasites derive nutrition from their host, thereby depriving the host of nutrients. In gaining their nutritional requirements, many parasites damage the host's tissues either by burrowing through them or by directly digesting them as a source of food. Multiple phyla contain parasitic members.

The **helminths**, or worms, are distributed over a number of phyla and each phylum contains some parasitic species. There are also a few members of the phylum Arthropoda that are parasites.

Helminths (worms)

Parasitic worms (helminths) of humans and other animals cause destructive diseases of major global socio-economic importance. In particular, parasitic flatworms (trematodes and cestodes) and roundworms (nematodes) have a massive, long-term impact on animal and human health, and cause substantial suffering, particularly in children. Figure 7.2.26 on the next page shows the current prevalence of worm infections and the proportion of those infected by country. As you can see, developing countries are most affected. Helminth infections are relatively uncommon in developed countries.

The World Health Organization (WHO) estimates that 2.9 billion people are infected with worms, mostly transferred from soil (soil-transmitted helminths, STHs) or from eating contaminated food. STHs are contracted from soil contaminated with faeces in areas where sanitation is poor. The incidence of diseases from worms surpasses diabetes and lung cancer in disability adjusted life years (**DALY**s).

Disability adjusted life years (DALYs) is a measure of the reduction in life expectancy due to ill health. It was devised so that differences in life expectancy in different countries due to disease burden could be compared.

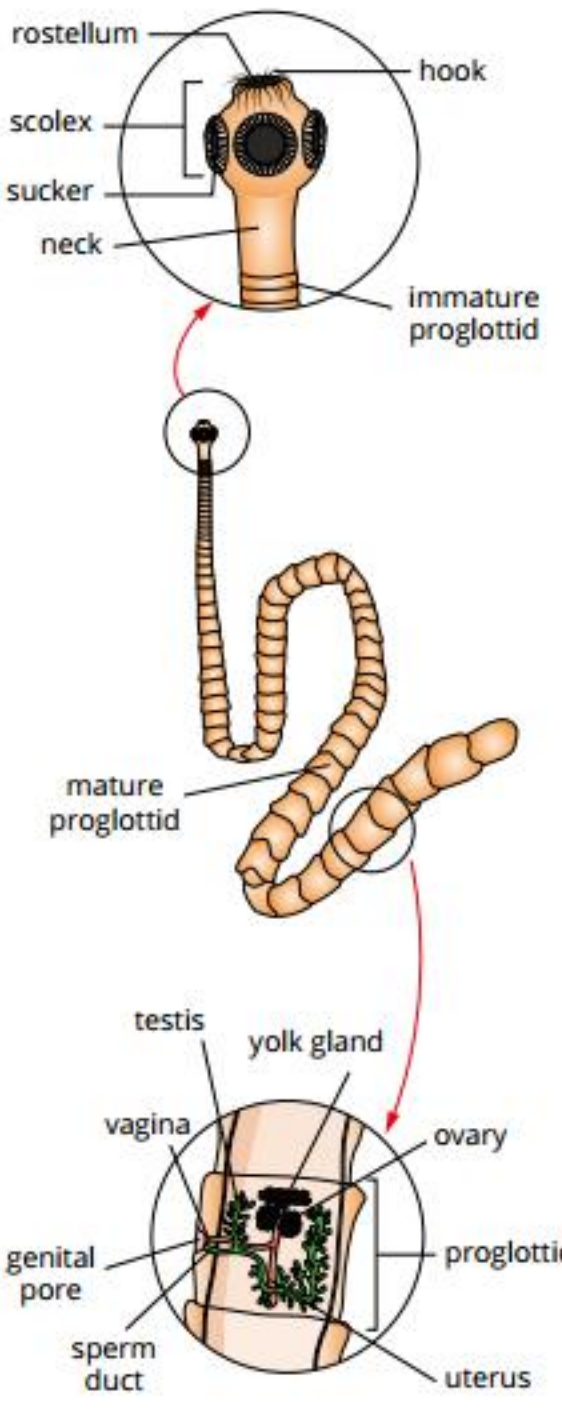


FIGURE 7.2.27 The structure of a tapeworm. Note the maturing segments near the tail that will break off as soon as maturation is complete. These segments are then able to infect a new host.

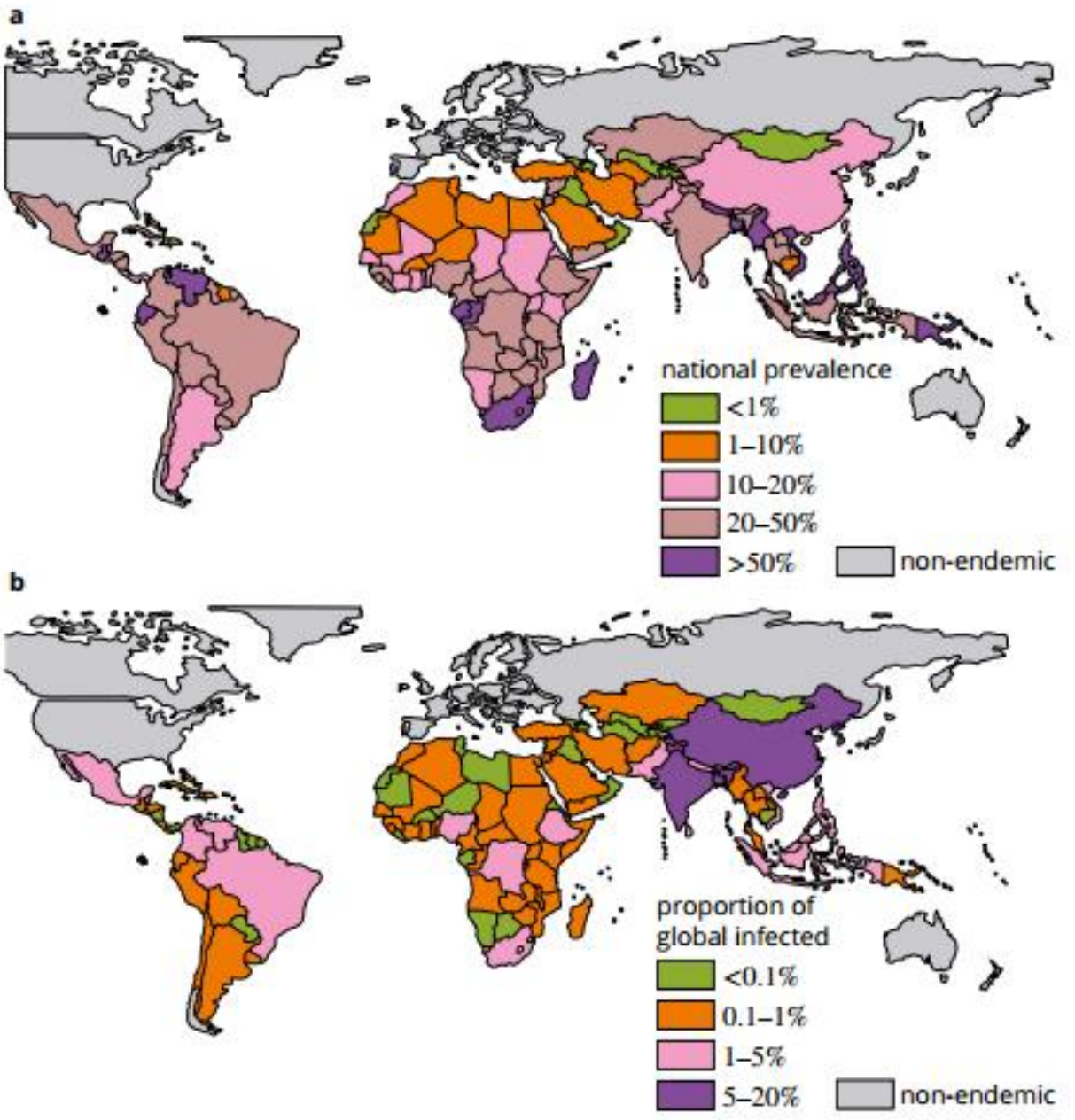


FIGURE 7.2.26 (a) Prevalence by country of infection with helminths and (b) the proportion of the world population infected living in each country. In Australia, helminths are non-endemic.

Parasitic worms include flatworms (phylum Platyhelminthes), such as tapeworms and blood flukes, and roundworms (phylum Nematoda), such as hookworms, dog heartworm, Ascaris and filarial nematodes (which cause elephantiasis). Helminths, which primarily inhabit the digestive tract, absorb nutrients, which can lead to malnutrition in the host. Other helminths consume blood from the host. A large enough infestation can result in anaemia from the depletion of the blood supply.

In plants, roundworms infect roots and are major pests of orchard trees and crops.

Platyhelminthes

Tapeworms are the most highly specialised parasitic flatworms. They are endoparasites that inhabit the gut of humans and other mammals. Adult worms may be up to 10 metres long and are flattened in shape. They have a head, which attaches to the wall of the gut, a neck region, which is the region of growth, and a chain of 'segments' (proglottids), which each contain a set of male and female reproductive organs (Figure 7.2.27). After fertilisation (usually between different segments), each segment matures into a bag of eggs that breaks off and passes out with the faeces.

The blood fluke, *Shistosoma mansoni*, is another platyhelminth known as a trematode. Its life cycle has a snail as the intermediate host (Figure 7.2.28).

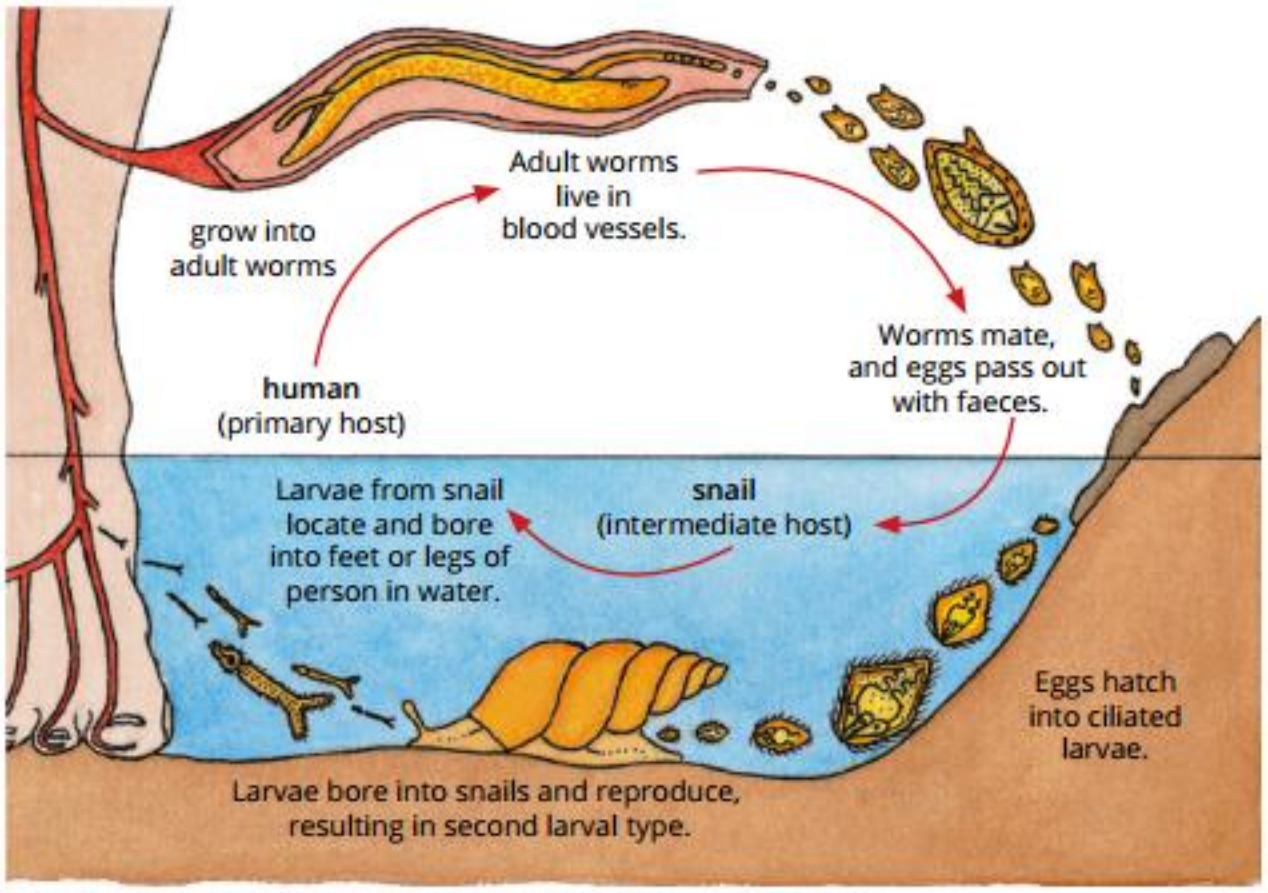


FIGURE 7.2.28 The life cycle of the blood fluke *Schistosoma mansoni*.

Nematodes

Nematodes are known commonly as roundworms, hookworms, threadworms or pinworms. They are present in huge numbers in the environment (millions in a shovel of earth; almost 100000 in a rotting apple) and are found anywhere that other organisms are found. Some species are extremely widespread while others have very specific requirements and are found in highly restricted habitats. Some species live in the guts of animals, such as dogs, as illustrated in Figure 7.2.29.

Although there are both free-living and parasitic nematodes ranging in size from microscopic to over 20 cm in length, the basic body structure of nematodes is virtually the same in every species. They have a simple body plan, lacking either a respiratory or a circulatory system. The pseudocoelom shown in Figure 7.2.30 is a fluid-filled cavity that bathes all the organs, allowing nutrient, waste and gas exchange to occur. Nematodes have a mouth and anus with a digestive tract that runs the length of their body. There are separate males and females.

FIGURE 7.2.29 The internal surface of a dog intestine infested with the hookworm, *Ankylostoma caninium*.

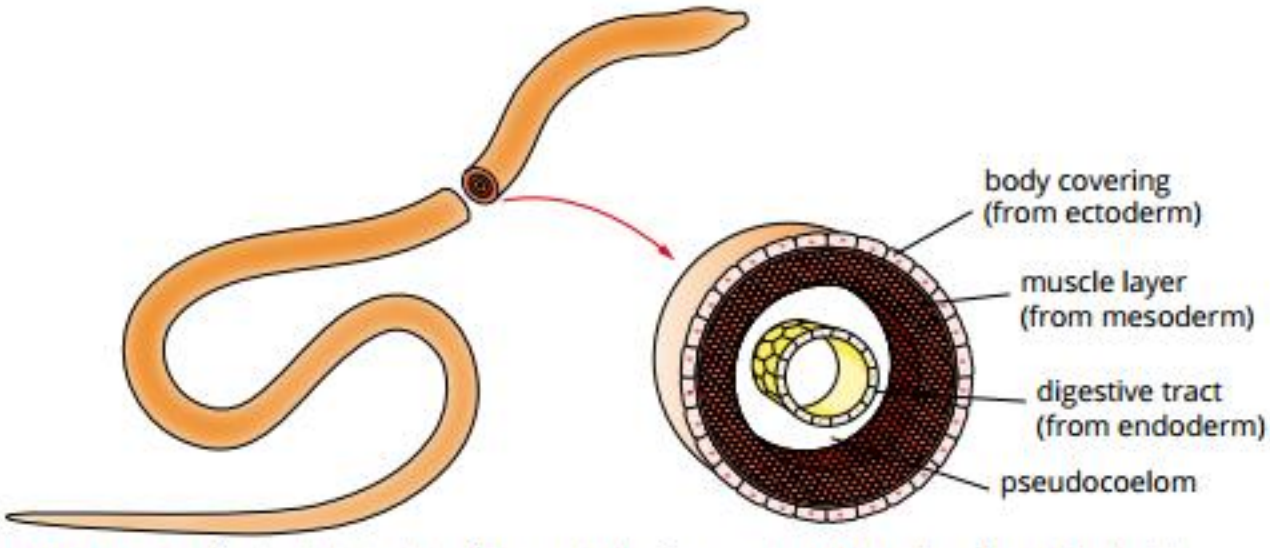


FIGURE 7.2.30 The basic body plan of a nematode. The pseudocoelom allows flow of materials throughout the body and negates the need for a circulatory or respiratory system.

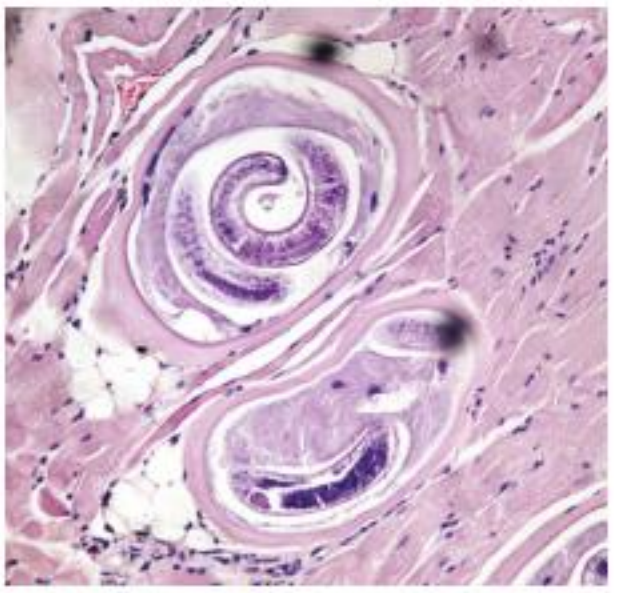


FIGURE 7.2.31 Light micrograph of cysts containing *Trichinella spiralis* worms in muscle tissue.

Parasitic nematodes include *Trichinella spiralis*, which causes a disease called **trichinosis** if uncooked, infested pork is eaten. Larvae in infected pork meat hatch out in the gut, where they develop into adult worms. While the adult worms cause no apparent harm and disappear after a few months, the next generation causes severe damage. Sexually mature adults in the gut produce millions of tiny larvae, which migrate through the wall of the intestine and into blood vessels. As they are carried throughout the body, the larvae leave the vessels and burrow into muscle tissues such as the diaphragm, rib muscles, tongue and eye muscles. Many of the symptoms of trichinosis are caused by mechanical damage of tissues as the larvae burrow through them, forming cysts that put pressure on the surrounding tissue. Figure 7.2.31 shows worm-containing cysts in muscle tissue. If cysts form in brain tissue, neurological symptoms can be observed. Fever is a common early symptom of infestation as inflammation reactions occur in response to the damage to body tissues.

Another nematode pathogen of humans is *Strongyloides steracolis*. It is endemic in tropical and sub-tropical areas, including areas of northern Australia, where it is particularly problematic in Indigenous communities. *Strongyloides* have two larval stages, **rhabditiform** and **filariform**. Rhabditiform larvae are excreted in faeces, infecting the soil, where they develop into either adults or filariform larvae. The main factor determining whether adults or filariform larvae eventuate is temperature. Lower temperatures (15–30°C) result in adults, while higher temperatures (>30°C) result in more filariforms. The life cycle of *Strongyloides* is illustrated in Figure 7.2.32. Note the two different pathways to infection.

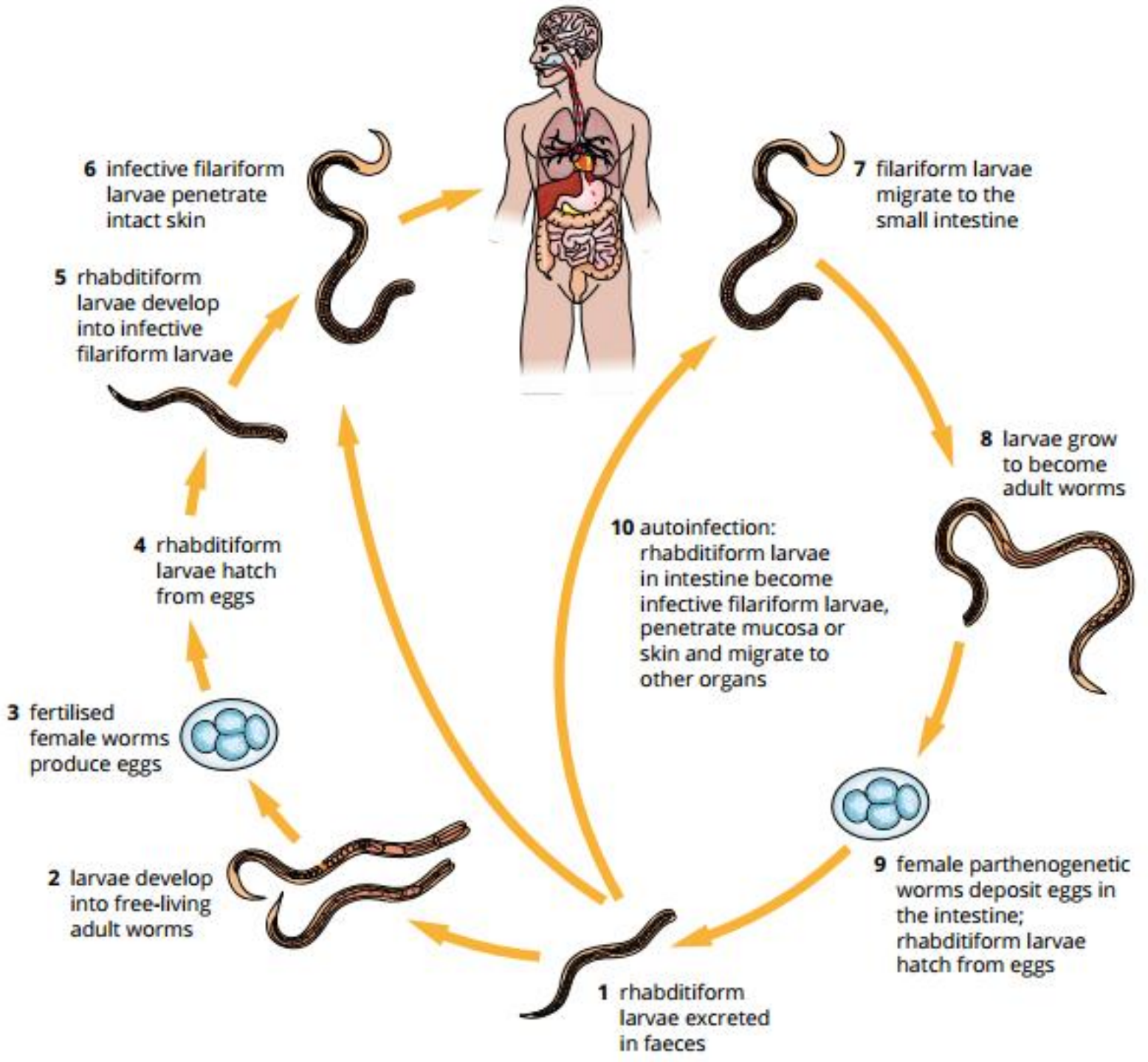

6 infective filariform larvae penetrate intact skin
5 rhabditiform larvae develop into infective filariform larvae
4 rhabditiform larvae hatch from eggs
3 fertilised female worms produce eggs
2 larvae develop into free-living adult worms
1 rhabditiform larvae excreted in faeces
7 filariform larvae migrate to the small intestine
8 larvae grow to become adult worms
9 female parthenogenetic worms deposit eggs in the intestine; rhabditiform larvae hatch from eggs
10 autoinfection: rhabditiform larvae in intestine become infective filariform larvae, penetrate mucosa or skin and migrate to other organs

FIGURE 7.2.32 The life cycle of the parasite *Strongyloides*.

Infection occurs when the filariform larvae penetrate the skin, after which they migrate in the blood to the lungs to be coughed up so that they can enter the digestive tract. The filariform larvae mature into adults in the small intestine. Abdominal pain is a common symptom, mainly caused by normal gut flora invading the surrounding tissues through damaged intestinal lining as the larvae burrow through the walls. Infection with gut bacteria and the presence of foreign nematode antigens in the tissues trigger an immune response characterised by a rise in body temperature (fever). If larvae migrate into the lungs, their burrowing causes physical damage to the tissue, resulting in breathing difficulties and chest pain. The adults release eggs, which hatch into larvae and are excreted in the faeces, further contaminating the soil. Spread of *Strongyloides* is common in areas where sanitation is poor.

Some nematodes are major pathogens of crop plants. Plant-infecting nematodes mainly attack roots, and part of their life cycle often takes place in the soil. Female worms lay their eggs in or on host tissues. The eggs hatch and the juveniles feed on nutrients obtained from host tissues, often stimulating tissue growth and gall formation. Nematodes that are parasitic on plants have distinctive mouthparts, with a sharp stylet that is used to puncture the cell wall (Figure 7.2.33). Many nematodes are also vectors (carriers) of other plant pathogens, including fungi, bacteria, and viruses.

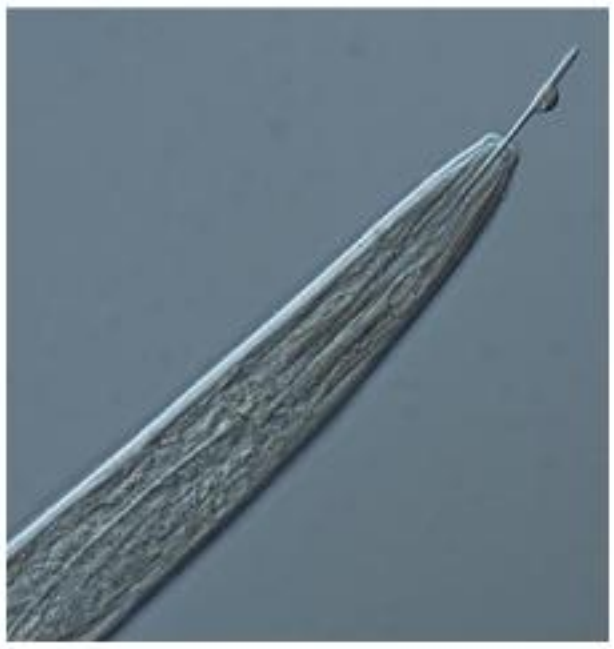


FIGURE 7.2.33 The mouthparts of many parasitic plant nematodes have a distinctive, needle-shaped stylus that is used to puncture the cell walls of the cells. The stylus is clearly visible in this light micrograph.

Arthropods

Insects are major vectors of disease in plants and animals. A few species of insects are parasitic on mammals, including some mosquitoes and fleas. Ticks and mites, including head lice, body lice and the crab louse, are examples of arthropods that infect humans.

In plants, **psyllids** (lerp insects) are small insects that, in their larval stages, induce the formation of galls on leaves (Figure 7.2.34). The larval stages of many psyllid species construct a covering (a lerp) under which they feed on the leaf surface. The saliva from feeding psyllids kills leaf tissue, causing extensive discolouration of the leaf. The saliva of the psyllid or damage to the plant tissue initiates increased production of plant growth hormones. This causes the plant's cells to grow larger or proliferate more quickly. In either case, the tumour-like gall surrounds the pathogen in a swollen mass of cells.

Other species of insects, including aphids, flies, wasps, and thrips, also induce the formation of galls on leaves and stems. The shape of the gall indicates the species of insect causing it. Sometimes a second parasitic insect lays its eggs into a gall. The new parasite kills the gall insect as it grows.

FIGURE 7.2.34 Psyllid-induced galls on a lilly pilly leaf

7.2 Review

SUMMARY

- Not all microorganisms can cause disease. Those that can are called pathogens.
- Infectious diseases are spread by pathogens being transmitted from one organism to another organism.
- Transmission can occur through direct touch or contact with body fluids, by inhaling or ingesting contaminated water or food, through vectors, or through medical procedures.
- Insects are vectors for many animal diseases and some cause galls in plants.
- Mosquitoes are one of the most important vectors for animal diseases.
- Virulence is the severity of disease caused by a pathogen. It depends on the characteristics of the pathogen and the host.
- Bacteria are classified according to their traits, which include structure, biochemistry and Gram staining.
- Many parasitic pathogens have complicated life cycles involving more than one host.
- Fungi and oomycetes are rarely important animal pathogens but are serious plant pathogens, sometimes destroying large areas of crops.
- Helminths (worms) are important human parasites, which can cause serious disease in some individuals.

KEY QUESTIONS

Describe

1 Identify four main methods by which diseases can be spread.

2 Describe how an infection with *Phytophthora* causes plant death.

3 Define 'gall' and explain the circumstances under which plants form them.

4 What are bacterial capsules primarily composed of?

5 What is a biofilm?

6 What advantages can biofilms provide to microbes?

7 **a** Name a fungus that causes disease in:
- **i** animals
- **ii** plants.

b Describe how the fungi in part **a** cause disease in their hosts.

Apply

8 Explain whether all bacteria are pathogenic. Use at least one example to clarify your answer.

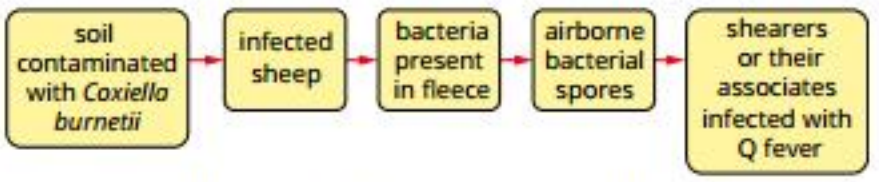
9 Describe how a primary host, an intermediate host and a vector are different.

10 How do adherence factors contribute to a pathogen's ability to invade and establish infection in a host?

11 Explain why gram-positive and gram-negative bacteria stain differently and state the significance of this in treating bacterial infections.

Analyse

12 Q fever is an example of a zoonotic disease. It is caused by a bacterium called *Coxiella burnetii*. In most people it causes a flu-like illness, but in some individuals it can cause liver or heart disease. One pathway for infection is shown here.

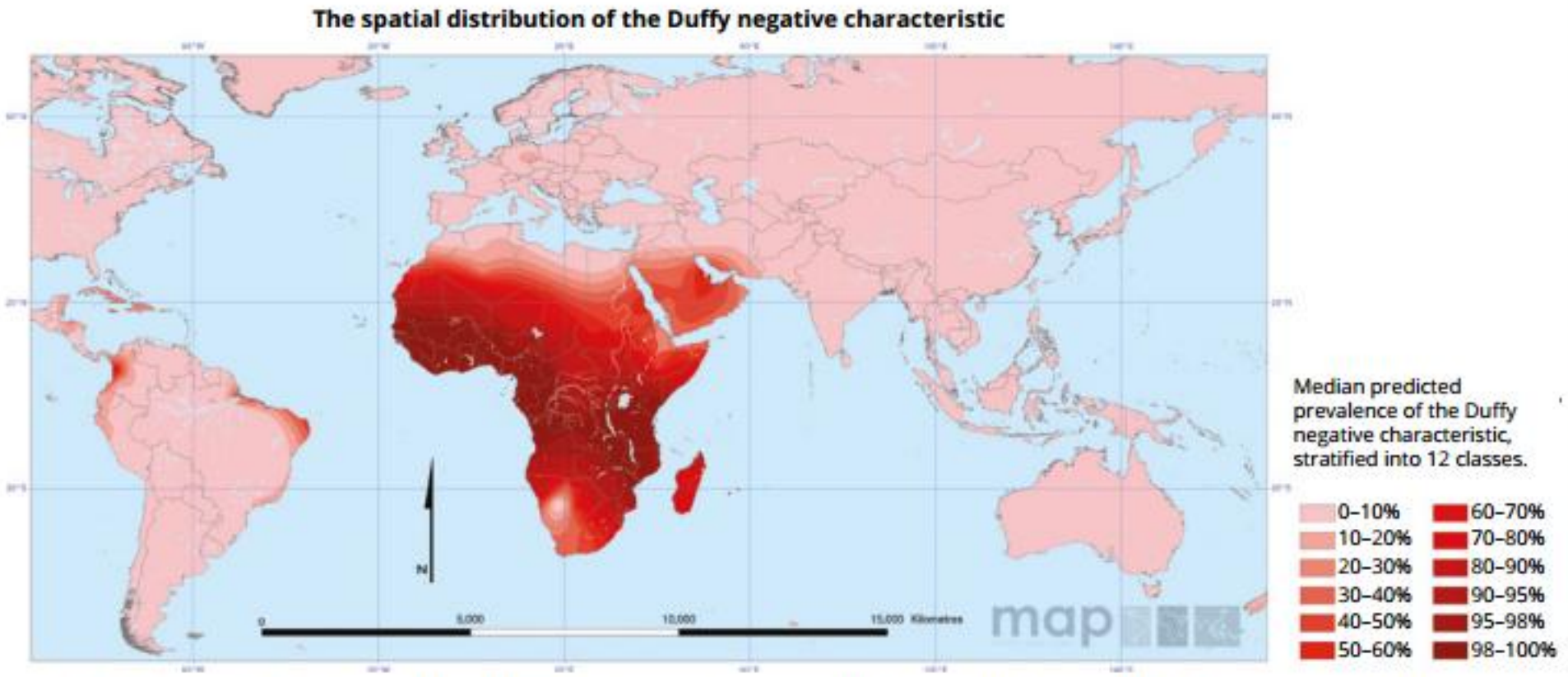

soil contaminated with *Coxiella burnetii*
infected sheep
bacteria present in fleece
airborne bacterial spores
shearers or their associates infected with Q fever

Propose three practices that could reduce the chance of infection with the bacterium.

13 Consider the life cycle of the malarial parasite (*Plasmodium*), which involves two hosts, as shown in Figure 7.2.13, page 281. Propose intervention strategies to stop disease or symptoms in humans.

14 Red blood cells carry surface molecules (antigens) for different blood groups, such as the ABO, MN, Rh and Duffy groups. Malaria merozoites of *Plasmodium vivax* use the Duffy antigen to recognise and attach to red blood cells. Individuals without Duffy antigens (Duffy negative) are at an advantage in regions where malaria is prevalent because the malaria parasite cannot easily enter their red blood cells. The map shows the distribution of the Duffy negative characteristic across the world.

Assess whether or not the data in Figure 7.2.12, page 280 and the map on the next page support the hypothesis that lacking Duffy antigen assists individuals to resist infection by *P. vivax*.



15 Poultry can be affected by ergot. Studies have been done on the effect of ergot on ducks that are bred for human consumption. The results of one such study is shown in the table below.

		Feed intake (g day^{-1})		Weight gain (g day^{-1})		Feed intake to weight gain ratio (g g^{-1})	
Group	Ergot intake (g kg^{-1} food)	Days 1–7	Days 8–14	Days 1–7	Days 8–14	Days 1–7	Days 8–14
A	0	35.1	98.5	25.7	66.6	1.4	
B	1	31.8	94.7	23.4	62.5		
C	10	25.4	78.7	17.2	54.4		
D	15	20.8	48.7	12.5	33.3		
E	20	18.7	38.1	10.7	24.8		

a i Calculate the ratio of feed intake to weight gain by the ducks and use your calculations to complete the table. (The first one has been done for you).

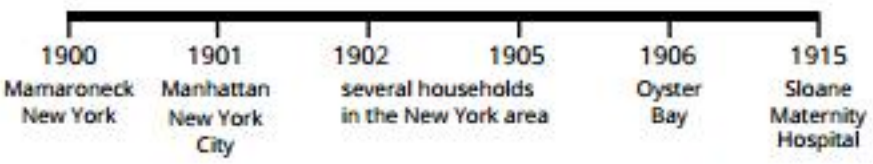
ii Explain what the value of 1.4 means.

b Explain which group is the control in this experiment.

c i Explain whether the claim, 'ergot causes a reduction in feed intake', is supported by the data.

ii Identify a second variable that influences feed intake.

16 *Salmonella typhi* is a bacterium responsible for the disease typhoid fever. Between 1900 and 1907, a woman named Mary Mallon was working as a cook in New York. Mary appeared to be perfectly healthy. A timeline of her working life is shown below.



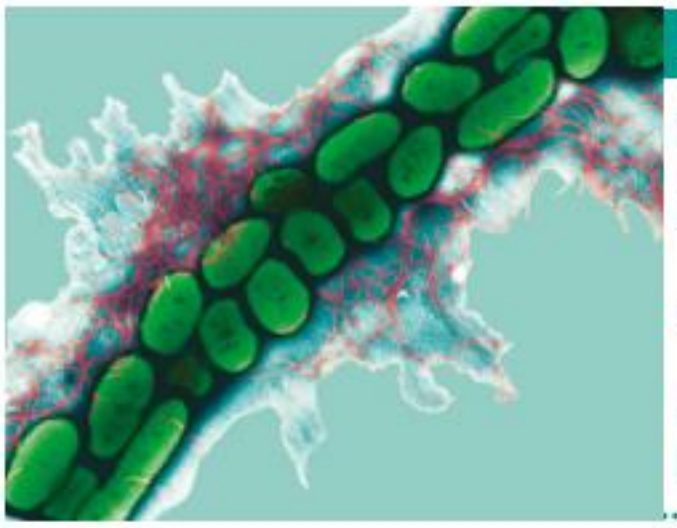
In 1900, Mary Mallon took a position as the cook in a series of private homes. Around 24 people became ill with typhoid. In 1901, she took a position in Manhattan; several members of the family, along with some of the other staff, became ill. From 1901 to 1906, she moved several times and at nearly all of the new positions, people became ill with typhoid. In 1906, she was working for a family in Oyster Bay on Long Island when 14 of the family became ill. For various reasons, Mary ceased cooking until 1915 when she took a position as cook at Sloane Maternity Hospital in New York. Twenty-five people became infected and two died.

a Explain how Mary could make others ill without showing any signs of disease.

b Explain the significance of Mary's occupation to the incidence of the typhoid outbreaks.

17 Pathogens of animals cause disease by many different pathways, but in plants the disease is often caused through similar mechanisms. Compare how *Agrobacterium* and *Fusarium* cause disease in plants.

7.3 Non-cellular pathogens



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that prions are abnormally folded proteins capable of causing other proteins to misfold
- explain how viruses use host cells for replication and then escape from the cell
- describe different viral structures and understand that bacteriophages are a type of virus that can only infect bacteria
- understand the role of host cell membrane receptors in viral entry into cells
- explain how antigenic drift and shift contribute to viral virulence.

Prions and viruses cause disease in many organisms. Unlike the cellular pathogens discussed in Module 7.2, prions and viruses are not made of cells, and so are known as non-cellular pathogens.

PRIONS

The smallest of all the known non-cellular pathogens are **prions**. Prions are the only known infectious agents that do not contain genetic material. Prions are proteins that are similar to normal cellular prion proteins (PrP), which are mainly located on the surface of the cells in the central nervous system of an organism. However, unlike PrPs, prions have an abnormal shape (or conformation) (Figure 7.3.1).

Prion diseases have two identifiable causes:

- infection
- genetics.

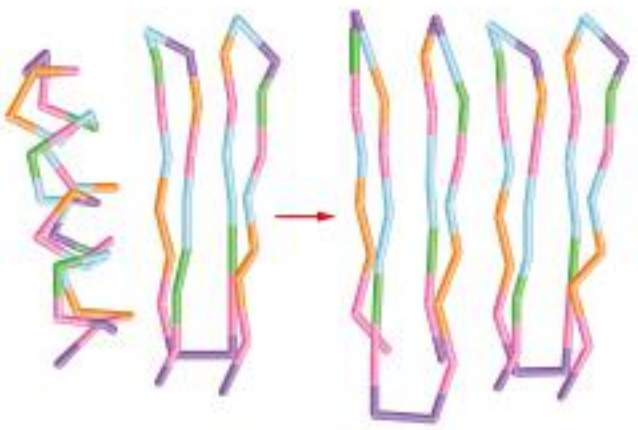


FIGURE 7.3.1 The normal spiral-shaped PrP protein unfolds and refolds into a pleated sheet shape. This makes the proteins clump together, or aggregate, to form masses called plaques, which damage neurons.

Infection accounts for 1% of the cases of prion disease, while genetics accounts for 10–15%. The remaining cases of prion disease (84–89%) are classified as sporadic, which means they have no clearly identifiable cause. Sporadic prion diseases may be caused by random mutations in single cells or by a random misfolding of a protein or another as yet unconsidered cause. Regardless of the source of the original misfolded protein, the progress of prion disease remains similar.

Prions stimulate the organism's normal PrP to misfold into the infectious prion form (Figure 7.3.1). The abnormal prion proteins maintain their ability to cause misfolding and thus continue to do damage. As a result of the mechanism by which prions cause disease, the damage to the nervous system develops at an exponential rate. By the time symptoms appear, which may be up to 30 years after infection, considerable damage has been done. The prion proteins build up in the tissues of the nervous system causing plaques. **Plaques** are masses of sticky proteins found in the brain where they lead to neuronal death by means that are still unclear.

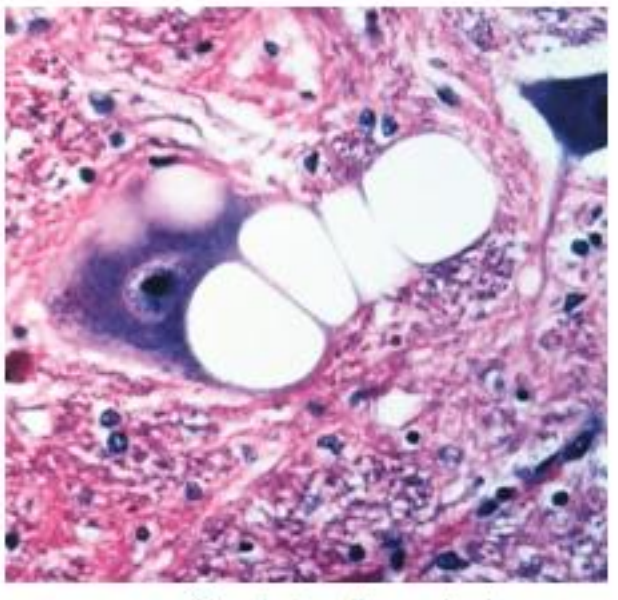


FIGURE 7.3.2 Scrapie is a disease in sheep caused by prions. This light micrograph shows a section through the brain of a sheep infected with scrapie. The large empty vacuoles (white) in the centre show the effects of the disease. As scrapie progresses, the number of empty vacuoles increases and makes the brain tissue appear spongy and destroys neurons. Symptoms of the disease include glazed eyes and body tremors.

Prions are resistant to being denatured, as well as to being broken down by proteases, which normally break down proteins in the digestive tract. Prions cause neurodegenerative diseases in mammals, the first of which to be discovered was scrapie in sheep.

In humans, prions cause **Creutzfeldt–Jakob disease (CJD)**. The CJD prions cause vacuoles and misfolded proteins (or plaques) to form in the brain, which kills neurons and makes the brain appear 'spongy' under a microscope. Figure 7.3.2 shows the typical spongy appearance of the brain tissue of a mammal, such as a sheep, affected by a prion disease.

Symptoms of CJD include dementia and sudden muscle contractions, leading to death. The equivalent disease in cattle, bovine spongiform encephalopathy (BSE), commonly known as mad cow disease, has been linked to a human variant CJD (vCJD) through human consumption of BSE-contaminated beef.

All forms of CJD have long incubation periods averaging around 10 years but there have been cases where onset of symptoms occurred more than 30 years from exposure. However, once symptoms appear, death generally occurs in less than 18 months.

The normal prion protein (PrP) has two common variants. One variant has the amino acid valine at the 129th position and the other has methionine in that position. The valine variant is designated V, while the methionine variant is designated M. Individuals can be homozygous (*VV* or *MM*) or heterozygous (*MV*). In caucasian populations, 50% of people are *MV*, 40% are *MM* and 10% are *VV*. It has been suggested that possession of the *MV* genotype partially protects against infection.

Prions elicit an ineffective innate immune response, and the adaptive immune system cannot identify and respond to them. You will learn more about the innate and adaptive immune systems in Chapter 8. It has been suggested that the reason the adaptive immune system cannot respond to prions is that, despite being abnormal in shape, prions remain very similar to the normal PrP, and any T-lymphocytes (a type of white blood cell) that would have responded to normal PrP would have been destroyed to prevent an autoimmune reaction. Another reason could be that prions are unable to be broken down and presented by antigen-presenting cells.

VIRUSES

Viruses are obligate, intracellular parasites, meaning they cannot replicate outside of cells. A single virus particle is called a **virion**. A virion is composed of genetic material, either DNA or RNA, enclosed in a protein coat, called a **capsid** (Figure 7.3.3). Some viruses also have a covering made of a combination of protein and lipid (a lipoprotein, shown in Figure 7.3.4c).

Viruses come in a variety of different shapes, but all have the same basic structure of genetic material enclosed in a protein coat. Figures 7.3.4 and 7.3.5 show some of the varieties of virion shapes.

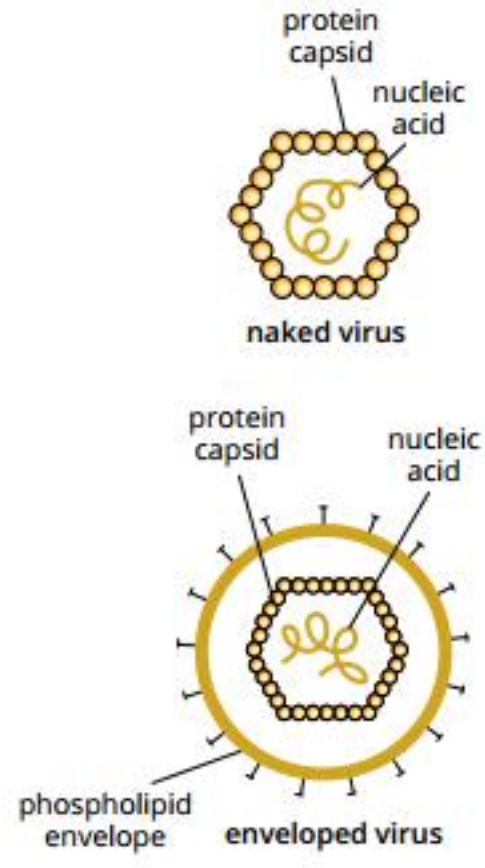


FIGURE 7.3.3 A virus is composed of a nucleic acid core, surrounded by protein molecules and, sometimes, a membrane envelope.

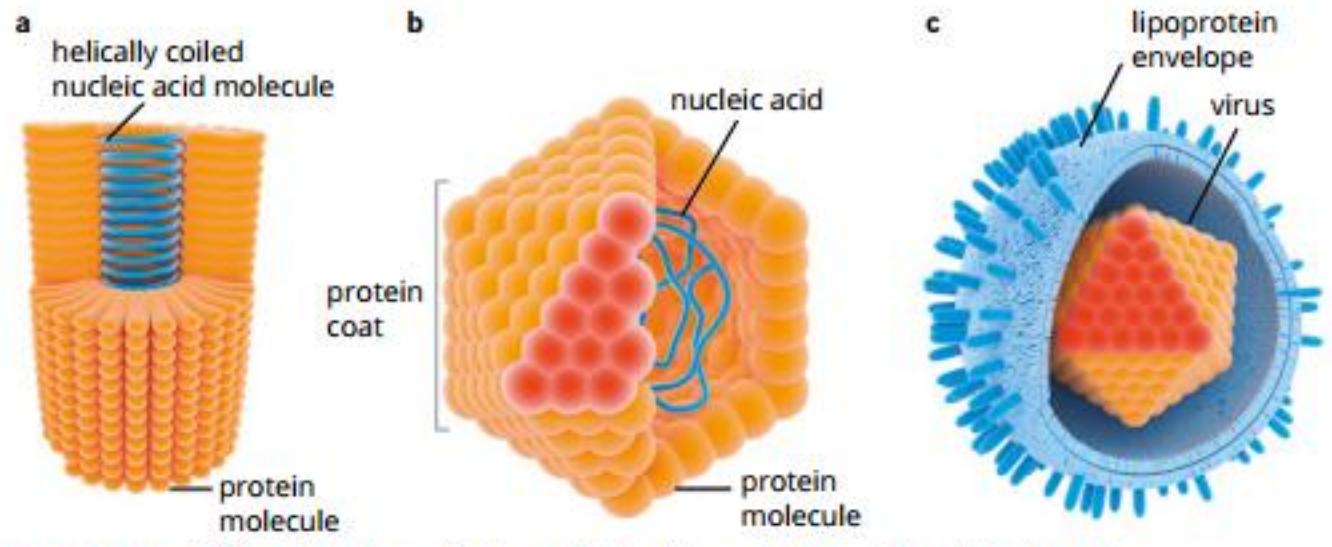


FIGURE 7.3.4 Different structures of virions. (a) A rod-shaped virion with proteins (orange) surrounding a helically coiled nucleic acid molecule (blue). (b) An isometric virion with an icosahedral protein coat surrounding a nucleic acid core (blue). (c) An icosahedral virion (orange) enclosed by a lipoprotein envelope (blue)

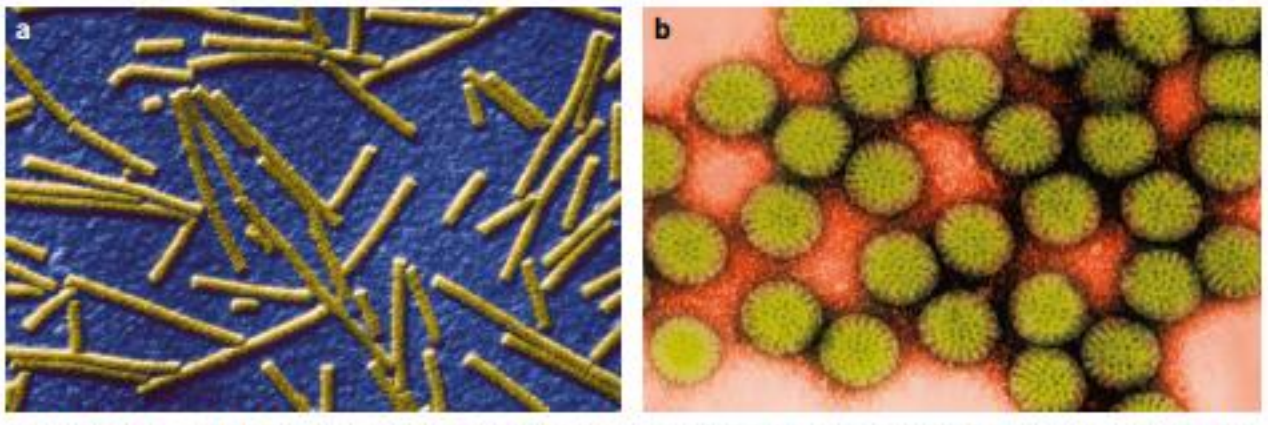


FIGURE 7.3.5 Coloured transmission electron micrographs of (a) a rod-shaped tobacco mosaic virus and (b) rotavirus, a major cause of gastroenteritis in children.

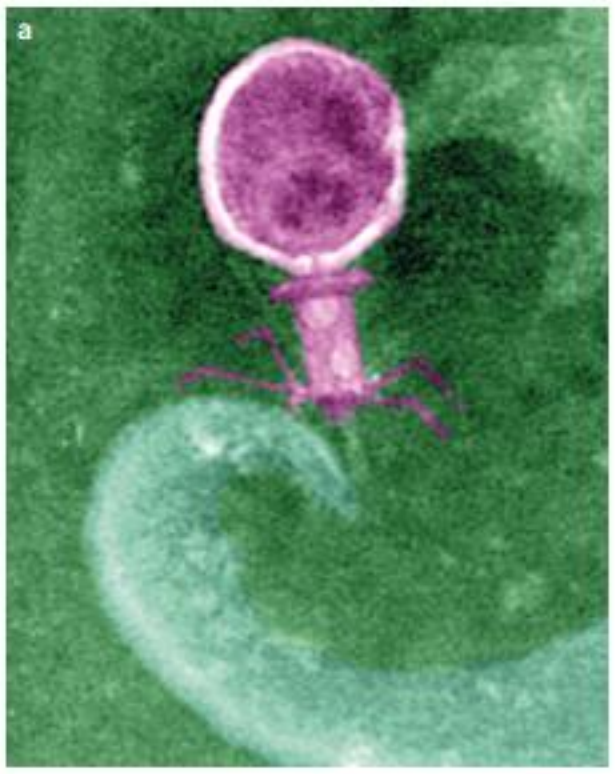


FIGURE 7.3.6 (a) A coloured transmission electron micrograph and (b) a diagram of a T2 bacteriophage. This is a virus that infects bacterial cells.

Viruses infect all types of living things, even bacteria. Viruses that infect bacteria are called **bacteriophages** (Figure 7.3.6). Bacteriophages cannot infect human cells and scientists are currently researching their therapeutic use for antibiotic resistant bacteria.

Viruses that use RNA as their genetic material are known as **retroviruses**. Retroviruses use an enzyme called **reverse transcriptase** to produce DNA, using their RNA as a template once they infect the host cell. HIV, which causes AIDS if untreated, is an important retrovirus.

The interaction between a virus and a host cell is specific. The specificity often lies in the attachment and entry processes, which involve viral proteins interacting with receptor molecules on the cell membranes of target cells. For example, the receptor for the influenza virus is a particular glycoprotein on the surface of red blood cells. In Figure 7.3.7, the virus is using one receptor on the cell to attach to the cell and a second receptor (called a co-receptor) to enter the cell by triggering endocytosis by the cell. Other viruses do not have specific attachment sites and enter cells by other means, such as engulfment by phagocytic cells. Once inside a cell, viral nucleic acid must survive the defensive enzymes of the host cell, which normally break up foreign DNA in order to prevent its replication. Some viruses have altered the structure of their DNA so that they are no longer susceptible to destruction by these host cell enzymes.

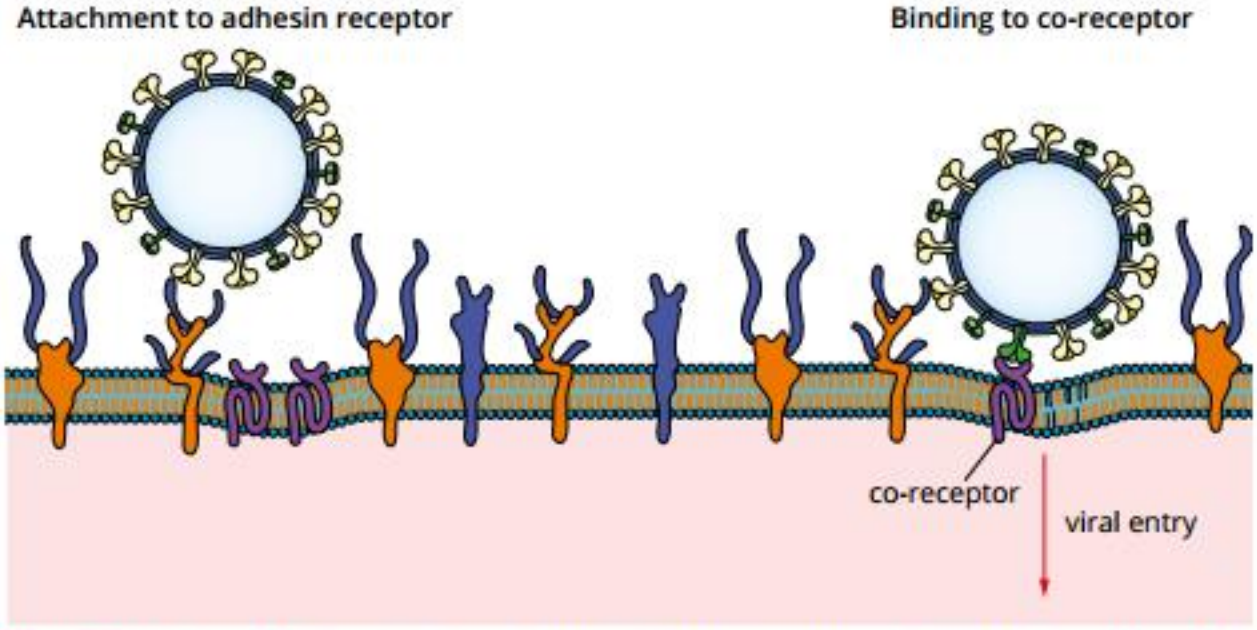


FIGURE 7.3.7 In order to infect a cell, viruses usually interact with receptor molecules on the outside of the cell membrane. Viruses may use a second receptor to assist with entry into the cell, called a co-receptor.

CASE STUDY 7.3.1

CCR5 and HIV

In 1995, it was observed that a group of people seemed to be highly resistant to infection by HIV. Untreated HIV results in immune suppression because it infects one of the crucial cells of the immune system called a helper T-lymphocyte. These white cells play a critical role in activating other cells of the adaptive immune response, so when they are invaded by HIV, the immune response is severely compromised and the ability to fight disease is decreased. You will learn more about the adaptive immune response in Chapter 8.

In order to enter the helper T-cell, the HIV virus attaches to a receptor on the outside of the cell called CD4. The HIV then uses another protein, called CCR5, to enter the helper T-cell. In most people, CCR5 is a large transmembrane protein that extends beyond the membrane both inside and outside the cell, and so can act as a co-receptor site. Figure 7.3.7 illustrates receptors and co-receptors.

Studies showed that some people have a mutation in the gene coding for the CCR5 protein, which makes the protein much smaller than normal. The mutated CCR5 protein is completely enclosed inside the membrane. This makes entry of HIV into the cells via CCR5 unavailable and, as a result, the people with this mutation are almost fully resistant to HIV. Individuals who are heterozygous for the mutation, while not fully resistant, have been shown to be much less likely to be infected by HIV and, if they are infected, the progression of the disease is much slower. One study examined the progression of HIV to AIDS in children who were infected from HIV-positive mothers during pregnancy. Data from this study showed a lower prevalence of infection in children with the CCR5 mutation and slower progression of the disease (Table 7.3.1).

If the virus evades the cell's defence mechanisms, it takes over the cell's metabolic machinery to produce new viruses. Instructions carried by viral nucleic acids direct the cell to produce viral proteins and nucleic acid, which are assembled into new virus particles. For DNA viruses and some RNA viruses, this production process is similar to normal protein production in cells. However, in retroviruses, including some cancer-related viruses and HIV, the process includes a unique step—the production of DNA from viral RNA. This is somewhat backwards because DNA is usually used to make RNA, not the reverse. Hence, the name 'retrovirus' because 'retro' means backwards. This backwards synthesis involves the action of reverse transcriptase. Figure 7.3.8a illustrates this process, using the example of HIV. This provides a window of opportunity for treatment, because a drug that selectively blocks this enzyme does not affect the normal functions of body cells.

Many viruses use the process of budding, which releases enveloped virus particles, occurs slowly and often does not result in the death of the infected cell. This results in a persistent infection of cells, as in the case of the herpes viruses, which cause cold sores and shingles, and HIV. Symptoms of herpes virus appear sporadically when the virus emerges from latency, such as when a person's general health is poor. Budding is shown in Figure 7.3.8b.

When viruses use budding as a means of escape from the host cell, the phospholipid bilayer of their membrane envelope comes from the host cell. However, the recognition molecules associated with it are of viral origin. Influenza viruses and HIV are also enclosed in such envelopes.

Other viruses build up inside the cell until they cause the cell to burst. The rupture of the cell allows the virions to escape and infect other cells. Infected cells that rupture to release the new virus particles are killed in the process.

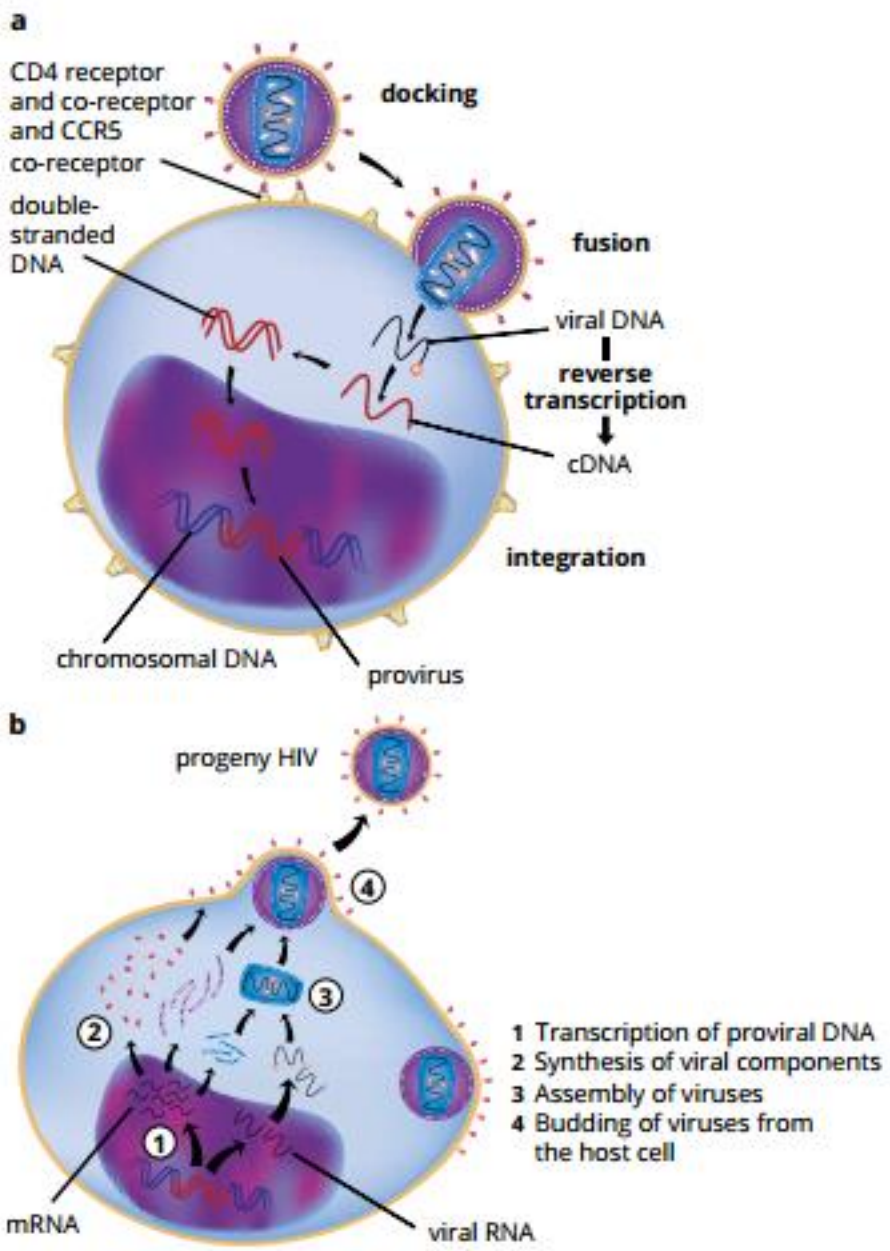


FIGURE 7.3.8 (a) HIV enters cells by attaching to a receptor called CD4. It uses a co-receptor called CCR5. When the viral RNA enters the cell, it is copied into DNA before it is incorporated into the nucleus of the host cell. (b) The host cell then makes new viral particles, which escape from the cell by budding.

TABLE 7.3.1 Progression to AIDS in children prenatally infected with HIV

CCR5 genotype	Speed of progression to AIDS		
	Rapid	Medium	Slow
homozygous normal	17	27	5
heterozygous	1	1	0
homozygous mutant CCR5	0	0	0
total cases	18	28	5

The importance of the CCR5 protein in allowing HIV to infect its host cells was further demonstrated by a case in Germany. In 2007, a patient infected with HIV and suffering from leukaemia was given a bone marrow transplant from a donor who was homozygous for the CCR5 mutation. Six months after the transplant, the recipient was tested for any evidence of HIV genetic material in his leukocytes and they were shown to be free of HIV. Eight years later in 2015, the bone marrow recipient remained free of both leukaemia and HIV.

Both the study of progression in children and the German case involved very small numbers of participants so any conclusions would need to be considered with caution. Further research is needed to determine if CCR5 mutations can hinder or halt the progression of HIV infection.

Review

1. Explain why results based on small studies should be treated with caution. Include natural variation and true value in your response (see Chapter 1, Part A).
2. From the evidence in Table 7.3.1., write a research question for further investigation.
3. Propose variables in humans that should be considered when conducting further testing, which would build the body of evidence and so the hypothesis can develop into a theory.

Antigens

Once a host has been infected, the host's immune system recognises antigens on the surface of the pathogen. Antigens are specific molecules, usually proteins or carbohydrates, which you will learn more about in Chapter 8. Once antigens have been identified, the host can mount an immune response. However, due to small random genetic changes, some viruses make minor changes to the antigens on their surface. This process is known as **antigenic drift** (Figure 7.3.9a). Usually, the changes in antigen structure are so minor that the host's immune system still recognises the virus (if a similar virus has previously infected the host). However, multiple episodes of antigenic drift over an extended period can result in significant changes in the viral antigens, effectively creating a 'new' virus. By contrast, **antigenic shift** (Figure 7.3.9b) is a much more abrupt change in the genetic code of a virus due to reassortment of genes from different viral strains, resulting in significantly different viral antigens. One source of antigenic shift is individuals who are simultaneously infected by two different strains of a particular virus. In this situation, the viruses can swap blocks of genetic material to give them new characteristics.

Antigenic shift can greatly increase the virulence of viruses. It is thought that antigenic shift was responsible for the worst influenza epidemic of all time, the Spanish flu. This influenza epidemic broke out in 1918 as World War I was coming to its end. Initially, because people were focused on the war, the flu did not attract much attention. However, by the end of the pandemic (a countrywide or worldwide disease outbreak; an epidemic is more localised), an estimated one-third of the world's population had been infected and approximately 50 million people (possibly up to 100 million) had died because of it.

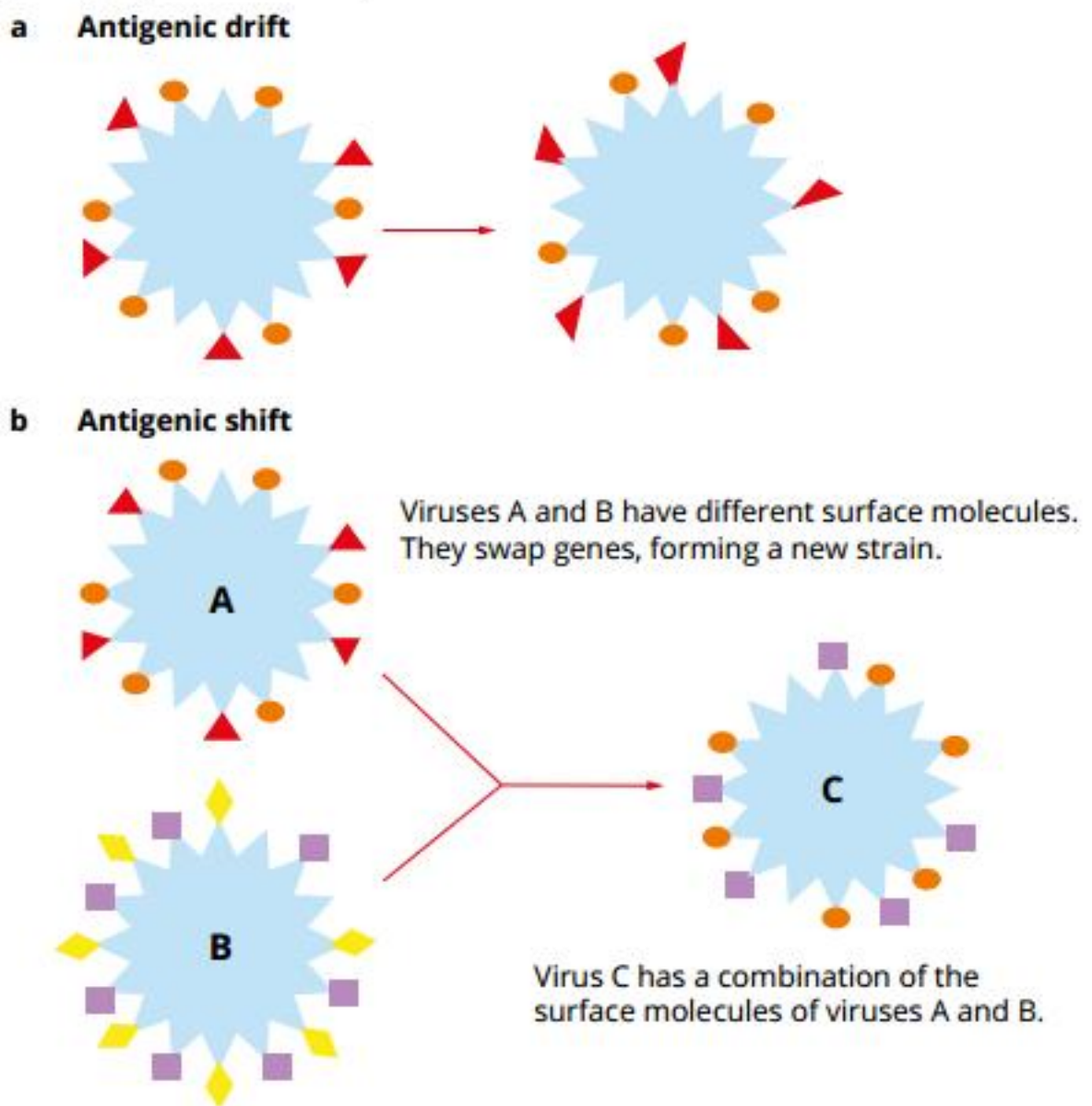

a Antigenic drift
b Antigenic shift
A
B
C
Viruses A and B have different surface molecules.
They swap genes, forming a new strain.
Virus C has a combination of the
surface molecules of viruses A and B.

FIGURE 7.3.9 (a) Antigenic drift is a slow process, which occurs as a result of the gradual accumulation of mutations. Initially, the differences are very small, but they accumulate over time. (b) Antigenic shift usually occurs as a result of gene swapping between viral strains and can result in significant changes in the surface molecules of a virus. It usually occurs when two different viruses infect the same host. They can then swap genes and gain very different surface molecules.

This is a mortality rate of between 10% and 20%. Some patients died directly from the influenza and others died from secondary infections, such as pneumonia, contracted while in a weakened state.

While there is no absolute certainty, genetic analysis of samples of the virus suggests that the Spanish flu was caused by a bird (avian) virus that gained the ability to pass from bird to human through antigenic shift. Antigenic shift can occur when humans and other animals live in close proximity.

Recent studies of the Spanish flu virus have given further insight into why it was so virulent. Researchers at the University of Alabama in the USA discovered that the virus contained a protein called NS1, which allowed it to attach to a human cell protein called RIG-1. The human protein RIG-1 is important because it is needed by the body to trigger an immune response to a viral invasion. By having a protein that binds to RIG-1, the Spanish flu virus could effectively avoid the activation of an immune response and thus replicate completely uncontrolled.

Plant viruses

Plant viruses cause diseases that affect many agricultural crops. There are three main groups: yellows, mosaics and necrotics. Yellow viruses tend to produce yellowing (and other damage to leaves and branches), mosaics produce mottling and spotting, and necrotics cause local or widespread death of plant cells. The vectors for plant viruses are insects, such as leaf-hoppers and aphids. Yellow viruses tend to persist in insects, whereas mosaics do not. Some viruses require an incubation period of a few days within an insect before they are infective, but what takes places during this period is not understood.

VIROIDS

Viroids are a type of self-cleaving RNA enzyme (or ribozyme) that is composed of short, circular strands of RNA that lack a protein coat. Viroids are only known to be pathogens of plants. They damage a plant by competing for nucleotides and forming viroid bundles, which mechanically interfere with the internal structure of the plant, much like a tumour. Viroids are known to have an unusually high mutation rate, which creates antigenic variation and allows them to avoid host resistance mechanisms.

Viroids are self-cleaving RNA enzymes (or ribozymes) only known to infect plants.

COMBATING PATHOGENS

Many pathogens, both organisms and molecules, rely on contact or poor sanitation to spread between hosts. This is mostly due to pathogens either being immobile or having limited mobility, relying on aid to spread from one host to another. Good hygiene is an important means of limiting the spread of most pathogens. This includes regular hand washing and bathing, proper disposal of contaminated materials, such as tissues, and sensible precautions to stop the spread of airborne particles, such as covering the mouth when coughing.

Transmission of body fluids between individuals is a mechanism for infection. Blood, semen, saliva and vaginal secretions are all fluids that can potentially pass pathogens between individuals. Where pathogens are spread by biting insects, insect repellents, bed netting and avoiding areas where the vectors congregate are helpful. You will learn more about mechanisms of transmission and combating disease as individuals, communities and globally in Chapter 9.

When infection has occurred, treatment is available for many of the cellular pathogens. Bacteria can be treated with **antibiotics**, which are chemicals that disrupt bacterial metabolism and/or their reproduction. Fungal infections and worm infestations in humans are generally treated with chemicals toxic to the parasite. There are treatments for some viruses, which slow their replication or disrupt their ability to escape from the host cell to infect other cells, but no treatments are available for prions.

CASE STUDY 7.3.2

Avian influenza 2003

Influenza viruses are normally classified by the variants of two proteins: **haemagglutinin** (H) and **neuraminidase** (N). Haemagglutinin is a surface protein of the virus that assists it to attach to potential host cells and gain entry. There are 18 known variants of this protein. Neuraminidase is an enzyme that allows the viral particles to escape from the host cell and there are nine variants of it. Influenza strains are named according to the variants of these two proteins. For example, the H1N3 strain has both variant 1 of haemagglutinin and variant 3 of neuraminidase.

In 2003, a H5N1 variant of avian flu crossed to humans and caused worldwide panic because mortality was so high (possibly 60%). Table 7.3.2 shows the cumulative number of confirmed human cases of H5N1, including deaths, between 2003–11. Scientists have postulated that as with the earlier Spanish flu, the H5N1 virus was particularly virulent because large parts of the population had not been exposed to any similar viruses. The viral strain that had been responsible for seasonal flu outbreaks for many years prior to 2003 had been either H1N1 or H3N2 strains.

It was initially thought that the H5N1 flu first crossed to humans in Vietnam because that was where the first large-scale outbreak was reported (Table 7.3.2). Common farming and poultry practices in Vietnam result in all or most family members coming into contact with the farmed animals. People in close contact with infected domestic birds were also infected. There was no evidence of person-to-person transmission at that stage.

More extensive studies showed that H5N1 had first appeared in China in wild birds and then in poultry.

After its initial evolution, H5N1 spread throughout Asia and Europe. There were also outbreaks in Africa. The virus was identified in several migratory bird flocks. It is thought that the virus was spread by migratory birds to local domestic birds, and then from the poultry to humans. Figure 7.3.10 shows the possible migratory paths of birds that may have carried the virus. Nearly all of the infected people had been in direct contact with sick birds or their faecal matter. There were a small number of cases of family members becoming infected possibly by person-to-person transmission, but this is not certain.

Viruses that infect other animals do not necessarily have the ability to infect humans, and even when they do, they will not necessarily spread from human to human. Widespread culling of infected birds and strict quarantine of infected people stopped the spread of the virus and averted a crisis. However, the potential for a virus of other animals crossing to humans and creating another pandemic of equal or worse severity than the Spanish flu of 1918 remains an ever-present danger, as the world was reminded when the COVID–19 pandemic began in late 2019.

Review

1 Propose why H5N1 was not seen in humans in Europe even though it occurred in both wild and domestic birds.

2 Explain why scientists cannot be certain that family groups caught the infection as a result of person-to-person contact.

TABLE 7.3.2 Cumulative number of confirmed human cases (C), including deaths (D), for avian influenza A (H5N1) reported to WHO, 2003–11 (laboratory cases only). All dates refer to onset of illness.

Country	2003		2004		2005		2006		2007		2008		2009		2010		2011		Total	
	C	D	C	D	C	D	C	D	C	D	C	D	C	D	C	D	C	D	C	D
Azerbaijan	0	0	0	0	0	0	8	5	0	0	0	0	0	0	0	0	0	0	**8**	**5**
Bangladesh	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	2	0	**3**	**0**
Cambodia	0	0	0	0	4	4	2	2	1	1	1	0	1	0	1	1	8	8	**18**	**16**
China	1	1	0	0	8	5	13	8	5	3	4	4	7	4	2	1	0	0	**40**	**26**
Djibouti	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	**1**	**0**
Egypt	0	0	0	0	0	0	18	10	25	9	8	4	39	4	29	13	32	12	**151**	**52**
Indonesia	0	0	0	0	20	13	55	45	42	37	24	20	21	19	9	7	7	5	**178**	**146**
Iraq	0	0	0	0	0	0	3	2	0	0	0	0	0	0	0	0	0	0	**3**	**2**
Lao People's Democratic Republic	0	0	0	0	0	0	0	0	2	2	0	0	0	0	0	0	0	0	**2**	**2**
Myanmar	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	**1**	**0**
Nigeria	0	0	0	0	0	0	0	0	1	1	1	0	0	0	0	0	0	0	**1**	**1**
Pakistan	0	0	0	0	0	0	0	0	3	1	0	0	0	0	0	0	0	0	**3**	**1**
Thailand	0	0	17	12	5	2	3	3	0	0	0	0	0	0	0	0	0	0	**25**	**17**
Turkey	0	0	0	0	0	0	12	4	0	0	0	0	0	0	0	0	0	0	**12**	**4**
Vietnam	3	3	29	20	61	19	0	0	8	5	6	5	5	5	7	2	0	0	**119**	**59**
Total	**4**	**4**	**46**	**32**	**98**	**43**	**115**	**79**	**88**	**59**	**45**	**33**	**73**	**32**	**48**	**24**	**49**	**25**	**565**	**331**

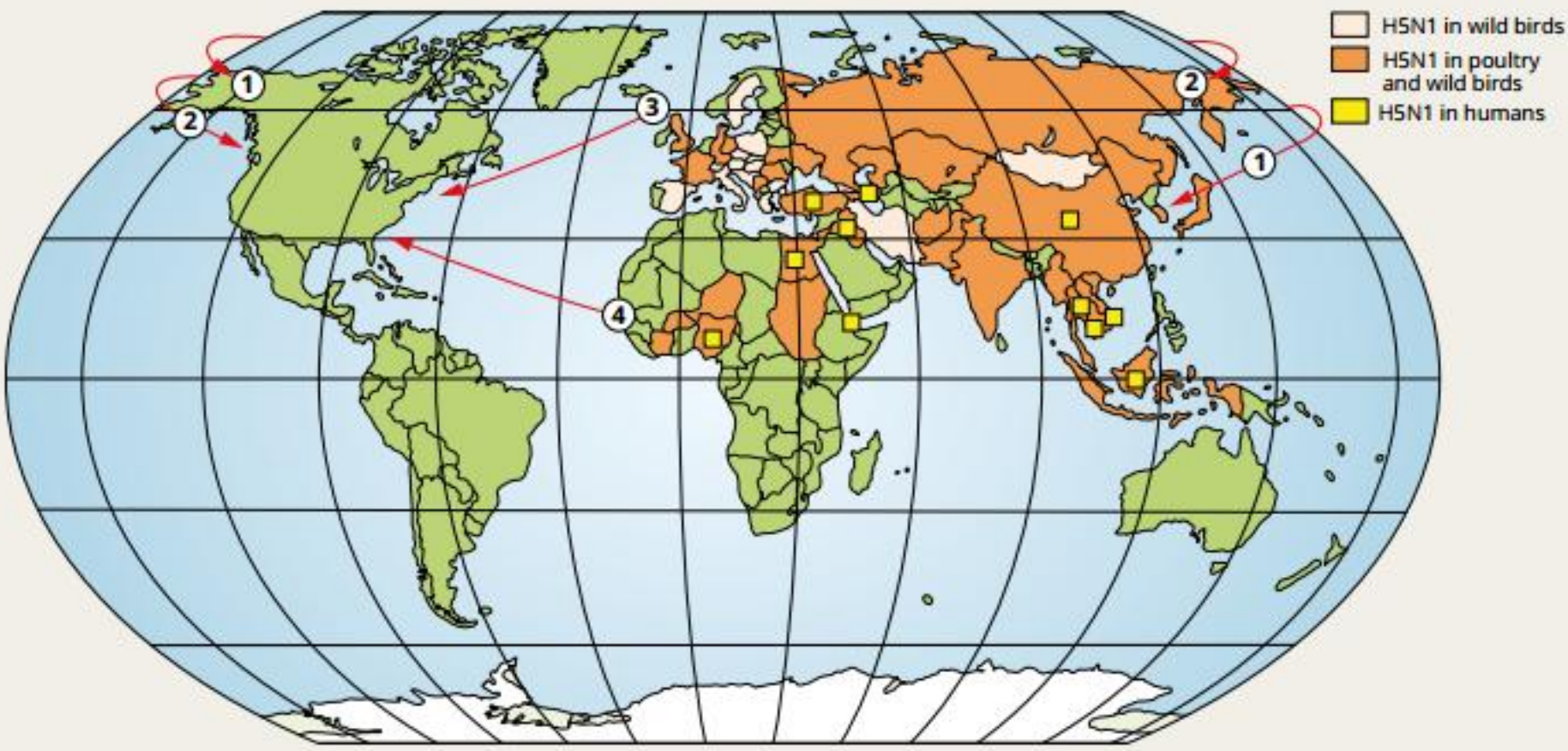


FIGURE 7.3.10 The actual and potential spread of the H5N1 strain of avian influenza A. Arrows indicate the known routes of wild bird migration. (1) Summer in Alaska, winter in East Asia. (2) Breeding in East Asia, winter in North America. (3) Breeding in Northern Europe and Iceland, winter in North America. (4) Birds carried by storms from West Africa to North America

7.3 Review

SUMMARY

- Prions and viruses are both non-cellular pathogens.
- Prion diseases, such as Creutzfeldt–Jabob disease (CJD), are caused by abnormally folded PrP proteins and are infectious because they cause other proteins to misfold.
- Some individuals have genetic traits that appear to give them resistance to prion infection.
- Viruses are infectious particles that consist of nucleic acids covered in a protein capsid.
- Some viruses use RNA rather than DNA. These are called retroviruses and include HIV.
- Viruses may escape from host cells by cell rupture or by budding.
- Genetics plays a role in determining the virulence of viral diseases. The CCR5 mutation in humans appears to offer protection from HIV infection.
- Viruses change over time, slowly by antigenic drift or quickly by antigenic shift. These changes can impact the severity of disease and the range of hosts able to be infected.
- Plant viruses fall into three main types: yellows, mosaics and necrotics.
- Plant viruses are often transmitted by insect vectors.

KEY QUESTIONS

Describe

1 Define:
- **a** bacteriophage
- **b** retrovirus
- **c** prion
- **d** antigen

2 Explain why the statement 'All viruses consist of DNA or RNA surrounded by a lipoprotein envelope' is false.

3 State two ways prions are different from viruses.

4 Identify the difference between an epidemic and a pandemic.

Apply

5 Explain why the incubation period of prion diseases is so long.

6 State how the PrP protein of an individual who has the *MV* genotype differs from that of someone who has the *VV* genotype.

7 Discuss the importance of vaccines in preventing viral infections compared to prion diseases.

8 How do viruses differ from prions in terms of their ability to reproduce?

9 Compare the genetic material of viruses, bacteriophages and prions.

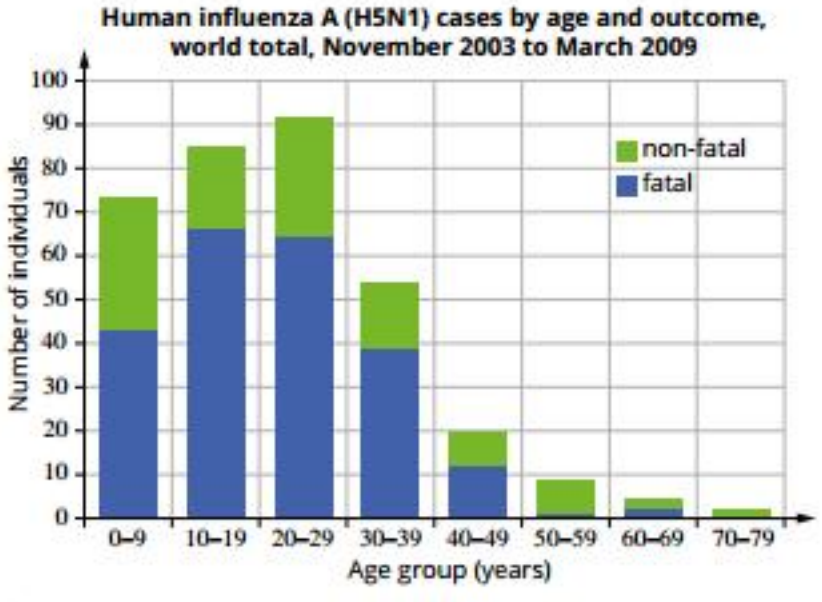
Analyse

10 Between the late 1950s and 1985, children who were identified as having conditions that would result in them being significantly shorter than average were treated with human growth hormone (HGH) to increase their height as adults. Until 1985, the source of HGH was the pituitary glands of human cadavers.

In 1985, the first case of Creutzfeldt–Jakob disease attributable to cadaver-derived HGH was diagnosed, leading to its use being stopped. Today, HGH is produced by genetically engineered bacteria. Up until 1985, approximately 30 000 people were treated with HGH; of those people, 139 developed CJD.

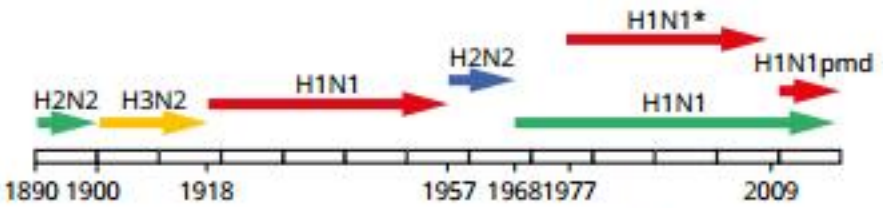
Calculate the chance of an individual treated with HGH from human cadavers subsequently developing CJD.

11 During the 2003 Avian flu (H5N1) outbreak, stringent quarantine precautions were taken. Not only were infected people and their contacts quarantined but in many countries including Australia, people arriving from affected countries were screened for any signs of fever and were quarantined if any was found. The reasons for these stringent precautions are suggested by the data in the graph, which shows global H5N1 cases by age and outcome, from November 2003 to March 2009.



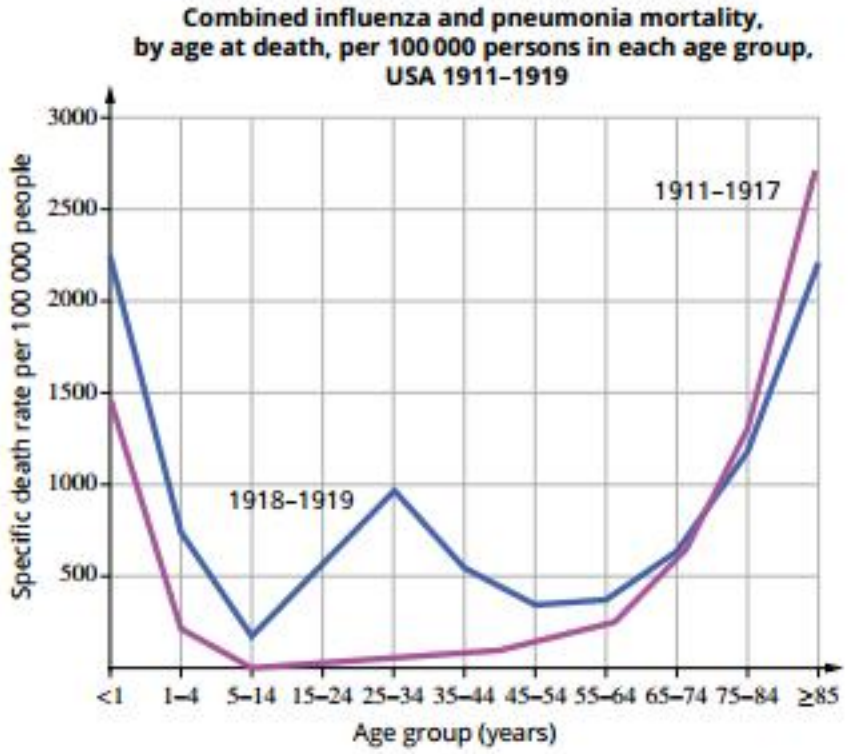
Consider why governments took such extreme precautions. Refer to the data in your answer.

12 Study of human tissues from past disease outbreaks has shown that waves of influenza have been with us for hundreds of years (at least). Some recent pandemics are shown in the diagram.



* Probably reintroduced from a laboratory from the H1N1 circulating from 1918 until 1957

Most influenza pandemics show a typical pattern of infection and mortality. The young and elderly are generally more likely to die. The following graph shows the fatality rate in the USA for the influenza pandemics of 1911–17 and for the Spanish flu pandemic of 1918–19.



The Spanish flu pandemic was very unusual in its distribution of fatalities. It has been hypothesised that the unusual pattern for the 1918 flu was due to older individuals having some level of immunity because of a previous influenza exposure and that people in their late teens and 20s had not been exposed to the relevant strain or anything like it. Consider the data in the diagram and the graph.

a Explain the evidence that supports this hypothesis.

b During the epidemic, death rates for 45–54-year-olds was low at around 375 per 10000. Extrapolate the death rate that would have occurred without the protection that 45–54-year-old people had.

13 Undertake independent research on the history of the use of bacteriophage to treat bacterial infections and provide a summary of why antibiotics became the preferred method for treating bacterial infections in 20th century, and why scientists are now once again researching the therapeutic use of bacteriophages.

Chapter review

07

KEY TERMS

adherence factor
adhesin
aeciospore
allele
antibiotic
antibody
antigen
antigenic drift
antigenic shift
auxin
bacteria (sing. bacterium)
bacteriophage
basidiospore
biofilm
capsid
capsule
carrier
chlamydospore
chromosome
contagious
Creutzfeldt–Jakob disease (CJD)
DALY (disability adjusted life years)
disease
Duffy
ectoparasite
endoparasite
endotoxin
epidemic
epidemiology
ergotism
exotoxin
filariform
fungus (pl. fungi)
gall
gram-negative
gram-positive
Gram stain
haemagglutinin
helminth
heterozygote advantage
heterozygous
homozygous
hypha
incubation period
infectious
infectious disease
intermediate host
karyotype
macrophage
malnutrition
merozoite
mutation
nematode
neuraminidase
non-infectious disease
oomycete
parasite
pathogen
pathogenicity
pathogenesis
peptidoglycan
pili
plaque
plasmodium
platyhelminth
primary host
prion
prostaglandin
protist
psyllid
retrovirus
reverse transcriptase
rhabditiform
toxin
trichinosis
vector
virion
viroid
virulence
virus
zoonotic disease
zoospore

REVIEW QUESTIONS

Describe

1 Kwashiorkor and scurvy are both deficiency diseases. Deficiency diseases are caused by:
- **A** viruses
- **B** bacteria
- **C** prions
- **D** malnutrition

2 It would be true to say of the bacterial capsule that it:
- **A** consists entirely of proteins
- **B** is necessary for bacteria to attach to a host
- **C** is important in allowing the bacterium to avoid phagocytosis by immune cells
- **D** is common to all bacteria

3 *Legionella* infection is caused by a bacterium that often contaminates air-conditioning cooling plants. The most likely method of disease spread of *Legionella* is:
- **A** vectors
- **B** airborne
- **C** body fluids
- **D** contaminated food

4 Haemagglutinin is important to the replication of influenza viruses because it plays an important role in:
- **A** attachment to the host cell
- **B** incorporating the DNA of the virus into the host's DNA
- **C** production of viral enzymes like neuraminidase
- **D** release from the host cell

5 Bacteriophage can:
- **A** only infect bacteria
- **B** only infect human cells
- **C** infect both bacteria and human cells
- **D** phagocytose viruses

6 Down syndrome is a genetic disease caused by having:
- **A** none of the number 21 chromosomes
- **B** only one of the number 21 chromosomes
- **C** two of the number 21 chromosomes
- **D** three of the number 21 chromosomes

7 Sickle cell-anaemia is a type of:
- **A** environmental disease
- **B** genetic disease
- **C** infectious disease
- **D** nutritional disease

8 The life cycle of the malaria parasite involves:
- **A** sexual reproduction in the vertebrate host, and asexual reproduction in the insect vector
- **B** asexual reproduction in the vertebrate host, and sexual reproduction in the insect vector
- **C** both asexual and sexual reproduction in the vertebrate host, and asexual reproduction in the insect vector
- **D** both asexual and sexual reproduction in the insect vector, and asexual reproduction in the vertebrate host

9 Define:
- **a** disease
- **b** pathogen
- **c** virus

10 List the ways in which disease-causing bacteria are spread.

Apply

11 Identify the incorrect statement about viruses.
- **A** They use the host cell for reproduction.
- **B** They are significantly smaller than bacteria.
- **C** They all contain DNA.
- **D** They consist of some nucleic acid enclosed in a protein coat.

12 Explain why lice are classed as ectoparasites.

13 Explain how capsules affect pathogenesis.

14 Discuss the importance of understanding capsule-mediated adherence in the context of combating bacterial infections. Provide examples of strategies or interventions that target bacterial capsules to prevent or treat infections.

15 Explain how bacterial toxins contribute to the pathogenesis of diseases like cholera and tetanus.

16 What are the main differences between endotoxins and exotoxins in terms of their origin, structure and effects on the host?

17 In a particular area, two strains of influenza virus have been circulating: H1N2 and H3N6. These influenzas have not caused major concern except for the elderly and in people whose immune systems are weak or compromised. Unexpectedly, there is a rapid increase in the number of young and previously healthy people presenting to hospital with influenza. A study of the virus shows it to be a H3N2 strain. This virus is likely to have arisen as a result of:
- **A** antigenic drift
- **B** antigenic shift
- **C** large numbers of people catching the virus from birds
- **D** a poorly targeted vaccination program

18 Compare pathogenic bacteria and fungi.

19 Explain the significance of both adhesins and pili in bacterial infection.

20 Compare viruses and prions.

Analyse

21 You are an astrobiologist on Mars and have discovered a microbe reminiscent of bacteria on Earth. You perform a Gram stain, and the organism stains pink. What conclusions can be derived from this information?

22 It has been observed that gram-negative bacteria are more likely than gram-positive bacteria to be involved in episodes of toxic shock (caused by endotoxins or exotoxins, or both), the most severe form of blood infection, in hospital intensive care units where patients are under specialised treatment. Explain this observation.

23 In a particular study, the chances of dying from a particular form of heart attack (myocardial infarction) between 1999 and 2008 had decreased by a probability of 0.82 in which the 95% CI range was 0.67–0.99. Explain what this means.

24 Studies of twins are important in determining the contributions of genetics and environment to disease. The following table shows the data collected from a series of studies in which pairs of identical and fraternal twins were examined for their body weight. BMI stands for body mass index and is calculated by dividing weight, in kilograms, by height, in metres squared. The column 'corr' is the correlation in BMIs between the two twins in each pair. It is a measure of how well the BMI of one twin matches the BMI of the other twin.

a Assess if a genetic component exists for BMI.

b i Calculate the standard deviation for the BMIs of female identical twins.

ii Males and females have different proportions of body fat and muscle. An equal volume of muscle weighs more than fat. Considering this information, propose why analysing male/female twin pairs is unlikely to provide useful information about the contributions of the effects of genetics and environment.

Adult identical twins						Adult fraternal twins								
Male/Male			Female/Female			Male/Male			Female/Female			Male/Female		
No. pairs	Mean BMI	Corr	No. pairs	Mean BMI	Corr	No. pairs	Mean BMI	Corr	No. pairs	Mean BMI	Corr	No. pairs	Mean BMI	Corr
1186	15.7	0.84	1429	15.40	0.84	1292	15.7	0.49	1204	15.50	0.50	2644	–	0.48
808	15.3	0.81	925	15.00	0.80	859	15.3	0.46	750	15.10	0.50	1633	–	0.45
474	15.0	0.79	544	14.80	0.88	497	15.0	0.64	437	14.90	0.57	947	–	0.48
630	15.2	0.88	738	15.30	0.88	645	15.3	0.58	580	15.40	0.54	1200	–	0.55
488	16.3	0.86	577	16.40	0.85	425	16.3	0.56	422	16.40	0.51	874	–	0.47
341	16.9	0.86	373	17.40	0.90	288	17.1	0.55	279	17.50	0.49	554	–	0.51
64	17.6	0.87	174	18.83	0.89	47	17.6	0.39	149	18.69	0.47	261	18.87	0.44
129	17.3	0.85	142	20.56	0.89	77	17.3	0.48	123	20.17	0.41	204	22.57	0.29
96	16.4	0.87	148	21.43	0.88	67	16.6	0.64	98	21.52	0.31	204	22.57	0.29
132	15.9	0.85	319	19.80	0.88	80	15.9	0.48	595	15.79	0.59	126	20.00	0.31
48	15.6	0.89	784	15.84	0.85	38	15.5	0.31	517	15.75	0.55	1010	15.79	0.54
120	15.4	0.83	720	15.64	0.81	67	15.5	0.45	593	17.26	0.52	819	15.75	0.45
139	15.3	0.91	804	17.08	0.88	95	15.5	0.55	735	17.83	0.48	950	17.26	0.45
110	15.5	0.91	823	17.75	0.86	71	15.7	0.35	354	17.60	0.50	1329	17.83	0.42
98	16.0	0.88	377	17.50	0.79	69	16.1	0.46	330	19.50	0.49	734	–	0.41
118	16.3	0.88	356	19.20	0.81	73	16.4	0.43	309	21.10	0.39	666	–	0.38
104	16.6	0.88	334	20.70	0.77	69	16.9	0.56	–	–	–	595	–	0.33
5085	**16.0**	**0.86**	**9567**	**17.57**	**0.85**	**4818**	**16.2**	**0.49**	**7475**	**17.50**	**0.49**	**14 750**	**18.83**	**0.43**

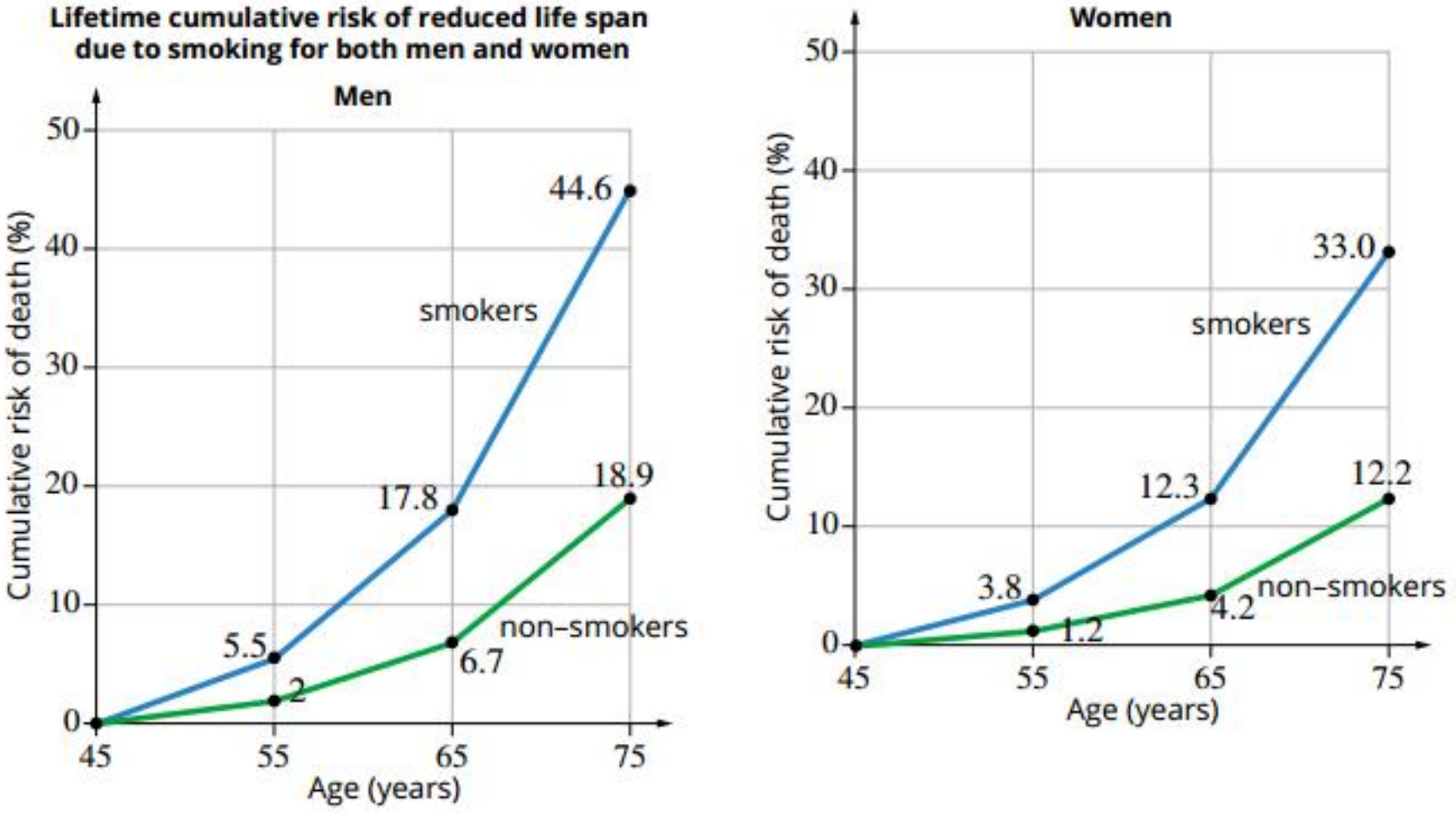
25 The lifetime cumulative risk of reduced life span due to smoking for both men and women is shown in the graphs on the next page.

a Determine the number of years a 65-year-old smoking female has lost, assuming that by comparing equivalent cumulative risk of death (%) between smokers and non-smokers, the number of years lost due to smoking is revealed.

b Calculate the difference in chance of death by age 75 of a male smoker compared to a non-smoker of the same age and sex.

c Determine whether the cancer mortality trends for Australia are worsening, on the basis of the data in the table below.

	Actual number of deaths	Rate (deaths per 100000 people)
1984	26645	212.3
1989	30555	214.4
1994	34134	212.6
1999	35575	194.2
2004	38489	184.6
2005	38836	181.6
2010	43216	196.2
2012	42961	189.0



26 Food allergies have become a significant source of disease in Australia in the last few decades. Many theories have been advanced to explain what seems to be a sudden and rapid increase. The following graph to the right shows the incidence of food allergies in school-aged children in a range of countries across the world. Data is not available for all countries.

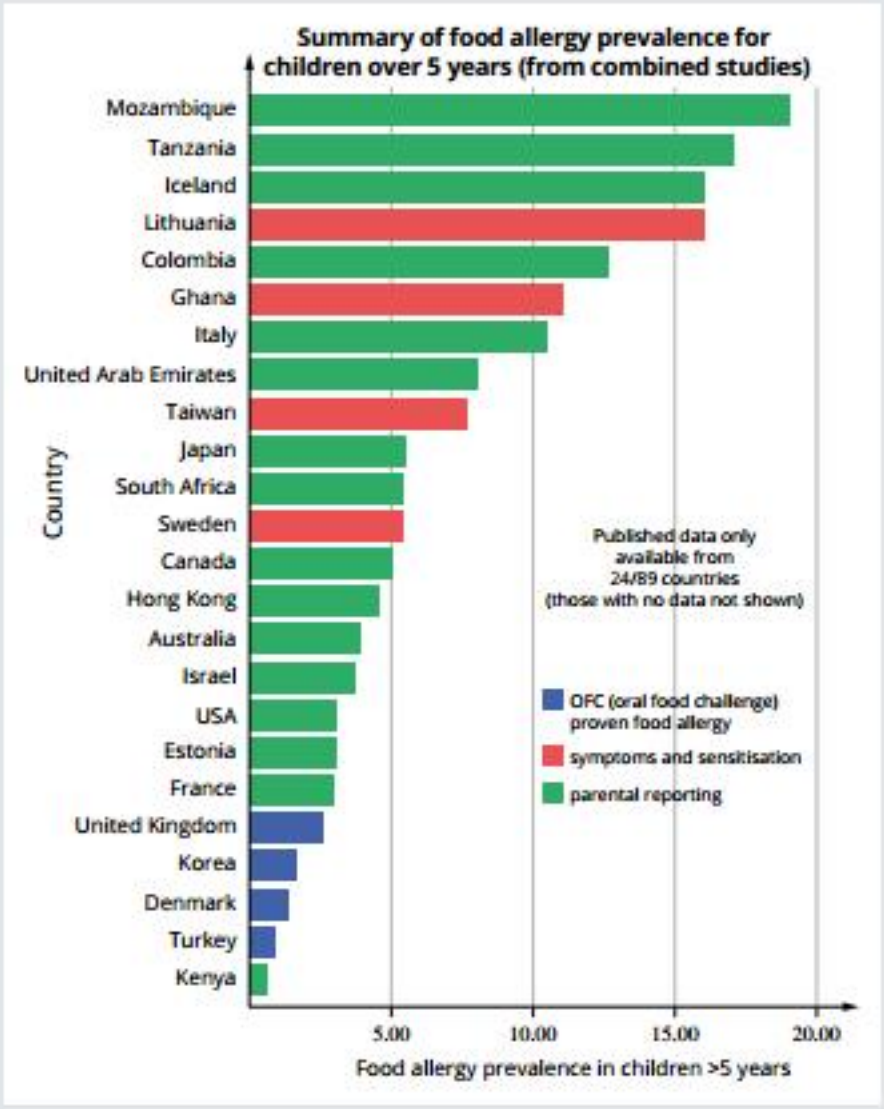
a Explain which of the two countries' data is likely to be the more reliable measure of the incidence of food allergies in their children, between the United Kingdom and Italy.

b The following data on food allergies was collected in the USA. Four thousand people were tested or interviewed for the survey.

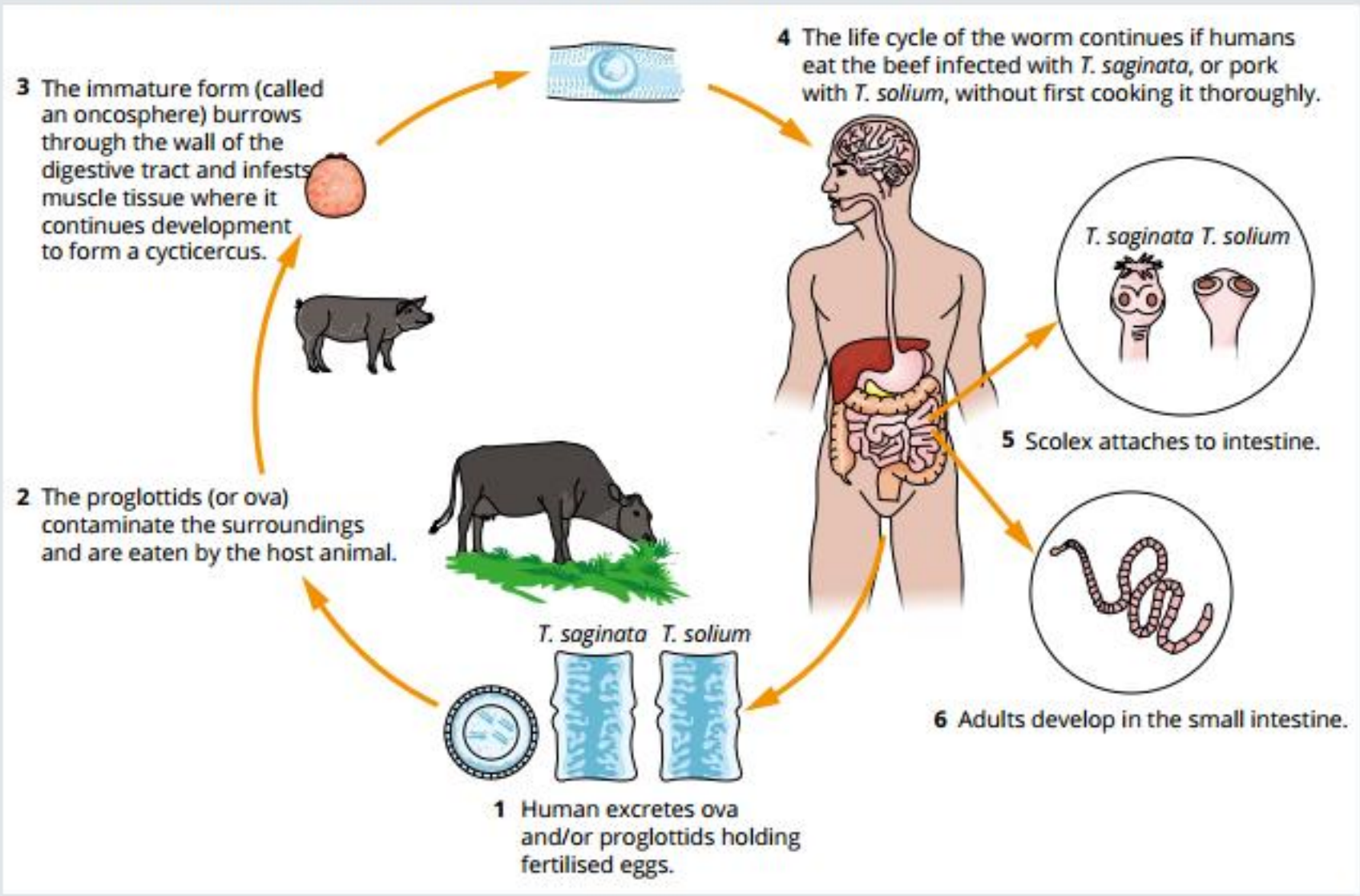
	Type of report	Year		
		2001	2006	2010
percentage of allergies reported by adults	self-reported	9.1	14.9	13
	medically diagnosed	5.3	7.6	6.5

i Discuss what the data suggests about the accuracy of self-reporting.

ii Explain whether there is any evidence that allergies among adults in the USA is increasing.



27 The life cycles of *Taenia saginata* (beef tapeworm) and *T. solium* (pork tapeworm) are shown below. Identify the primary and intermediate hosts, giving reasons for your decision.


4 The life cycle of the worm continues if humans eat the beef infected with T. saginata, or pork with T. solium, without first cooking it thoroughly.
3 The immature form (called an oncosphere) burrows through the wall of the digestive tract and infests muscle tissue where it continues development to form a cycticercus.
T. saginata T. solium
5 Scolex attaches to intestine.
2 The proglottids (or ova) contaminate the surroundings and are eaten by the host animal.
T. saginata T. solium
6 Adults develop in the small intestine.
1 Human excretes ova and/or proglottids holding fertilised eggs.

28 Argue which conditions are more likely to facilitate the spread of plant diseases caused by fungi and members of the oomycetes.

29 Many diseases, caused by both bacteria and viruses, have incubation periods ranging from several days to months or even years. Explain how this assists the pathogen to spread.

30 The following table shows data collected by WHO on H5N1 (bird flu) infection in wild and domestic birds and humans 2003–10. Numbers are for human cases. Coloured boxes represent animal outbreaks: light blue boxes represent sporadic and/or seasonal outbreaks notified in poultry; grey boxes represent poultry outbreaks reported throughout the year in domestic birds; white boxes represent no avian outbreak reported.

Country	Nov 2003–Dec 2005	2006	2007	2008	2009	To 1 July 2010	Total
Azerbaijan	0	8	0	0	0	0	**8**
Bangladesh	0	0	0	1	0	0	**1**
Cambodia	4	2	1	1	1	1	**10**
China	9	13	5	4	7	1	**39**
Djibouti	0	1	0	0	0	0	**1**
Egypt	0	18	25	8	39	19	**109**
Indonesia	20	55	42	24	21	4	**166**
Iraq	0	3	0	0	0	0	**3**
Laos	0	0	2	0	0	0	**2**
Myanmar	0	0	1	0	0	0	**1**
Nigeria	0	0	1	0	0	0	**1**
Pakistan	0	0	3	0	0	0	**3**
Thailand	22	3	0	0	0	0	**25**
Turkey	0	12	0	0	0	0	**12**
Vietnam*	93	0	8	6	5	7	**119**
Total	**148**	**115**	**88**	**44**	**73**	**32**	**500**

* With a high degree of seasonal and geographical variation
Source: Data collected by the epidemic intelligence team at Institut de Veille Sanitaire from postings on the websites of the World Health Organization, the World Organization for Animal Health and other authoritative sources in the 15 countries.

It has been proposed that bird flu spread from wild birds to domestic poultry and from domestic poultry to humans.

a Analyse the data in the table above and identify the evidence that supports the hypothesis that humans caught the flu from domestic poultry.

b The following table shows the distribution of deaths from H5N1 by age and sex.

Age group (years)	Male	Female
<5	13	8
5–9	19	13
10–19	18	31
20–29	18	27
30–39	17	16
40–49	5	6
>50	6	5

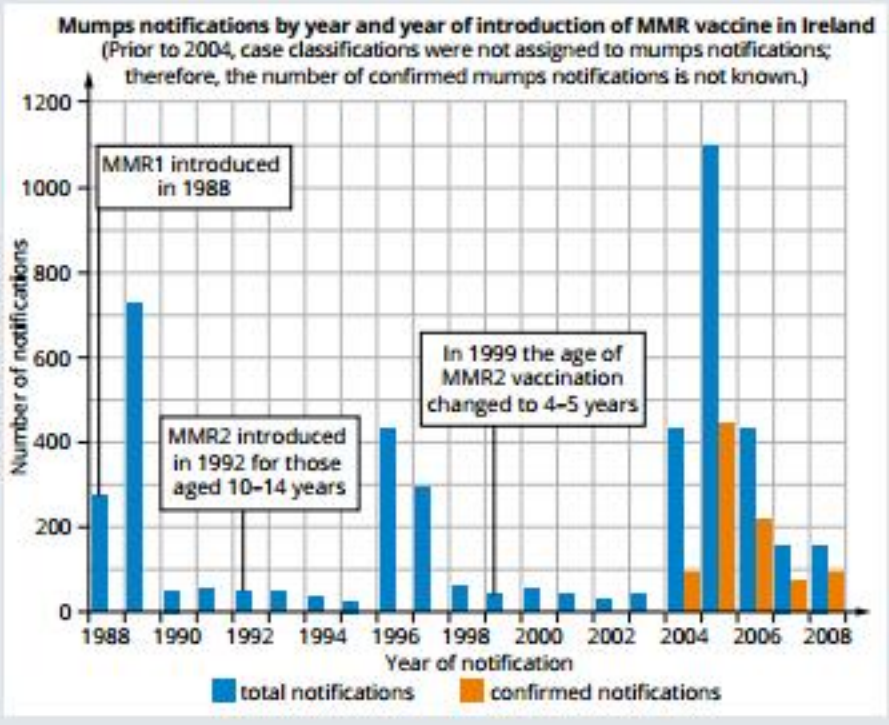
i Draw a graph of the data in the table.

ii The way the data is displayed is somewhat misleading. Explain how the data is misleading.

iii Explain whether the data suggests that either sex is more susceptible to bird flu.

iv The data given is from a WHO report. WHO does not collect data itself. It relies on self-reporting from affected countries. Explore the reliability of the data presented.

31 Mumps is caused by a virus. A vaccination (MMR) has been available since the early 1980s. The vaccination was introduced in Ireland in 1988. Once the vaccination was introduced, reporting of suspected cases became mandatory. The incidence of reported cases is shown in the following graph. From 2004, suspected cases were tested to confirm diagnosis. This did not take place before 2004.



a State the percentage of total notifications that were confirmed in 2006.

b Discuss the reliability of mandatory reporting of suspected cases, in consideration of testing to confirm mumps.

Interpret

32 In Australia, the prevalence of infection with threadworms (nematodes) is much higher in children under the age of 10 years than in adults. Argue why this is the case. Consider the life cycle of the nematode in your answer.

33 It has been suggested that the filarial forms of the helminth *Strongyloides stercoralis* are attracted to the potential host by chemicals in sweat called urocanic acid. Design an experiment to test this hypothesis. Identify the independent and dependent variables in your experiment and state what results would support the hypothesis.

34 **a** Discuss the nature of the HIV pandemic. In your answer, include the type of pathogen, its method of causing disease, its method of transmission, and its method of infecting tissues.

b Research HIV treatments and describe how different antiretroviral medications effectively manage HIV infection by targeting multiple stages of the HIV lifecycle.

35 Cholera is a serious disease caused by the bacterium *Vibrio cholera*. The disease is characterised by diarrhoea, vomiting and cramping. If untreated, severe dehydration and shock occur. Mortality can be as high as 50%.

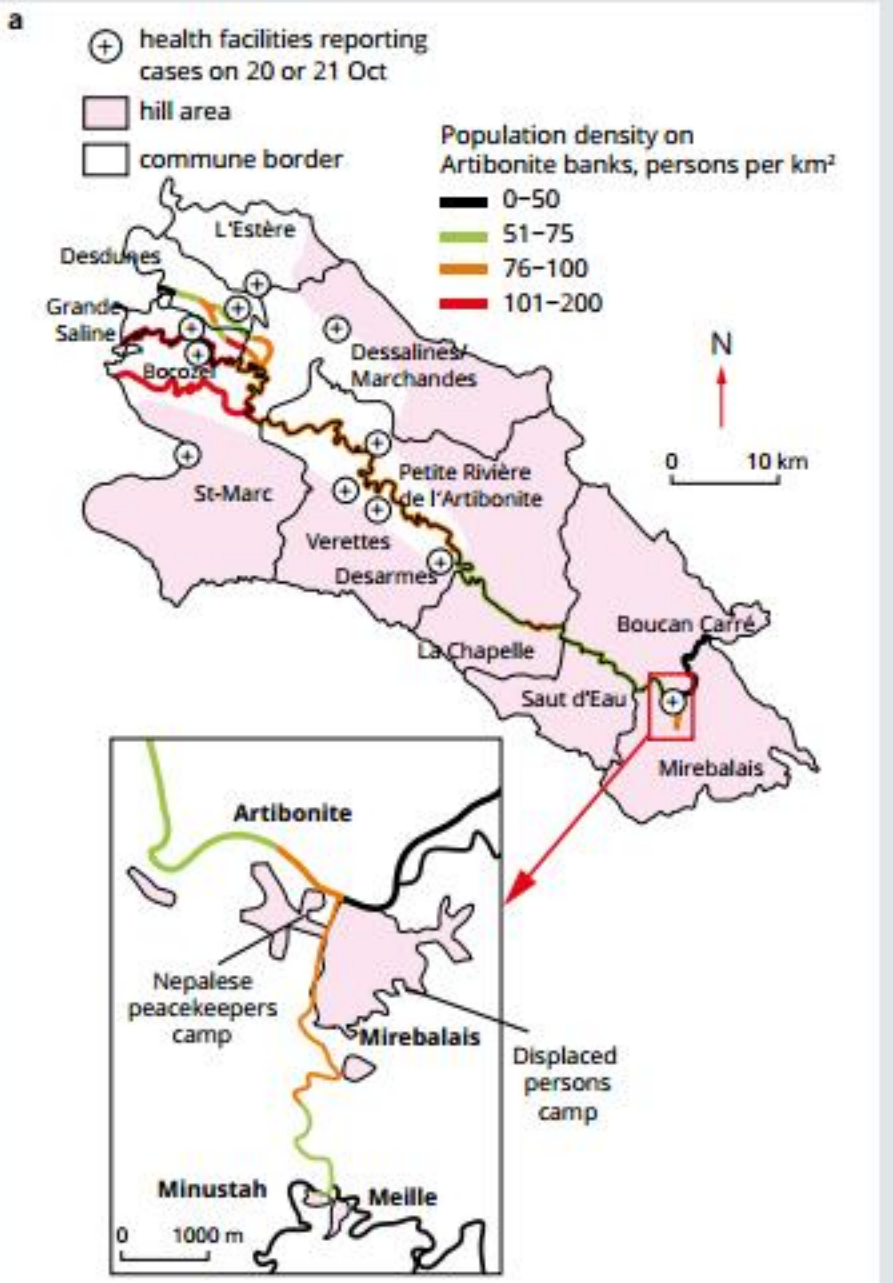
In 2010, the island of Haiti was shaken by a major earthquake that destroyed much of the infrastructure on the island, including electricity, sewage and water treatment plants. This left most people relying on untreated river water for all of their water needs. Many people were buried by falling buildings and so there was a large influx of people from many countries to assist, to find survivors, help with the provision of medical attention and maintain law and order.

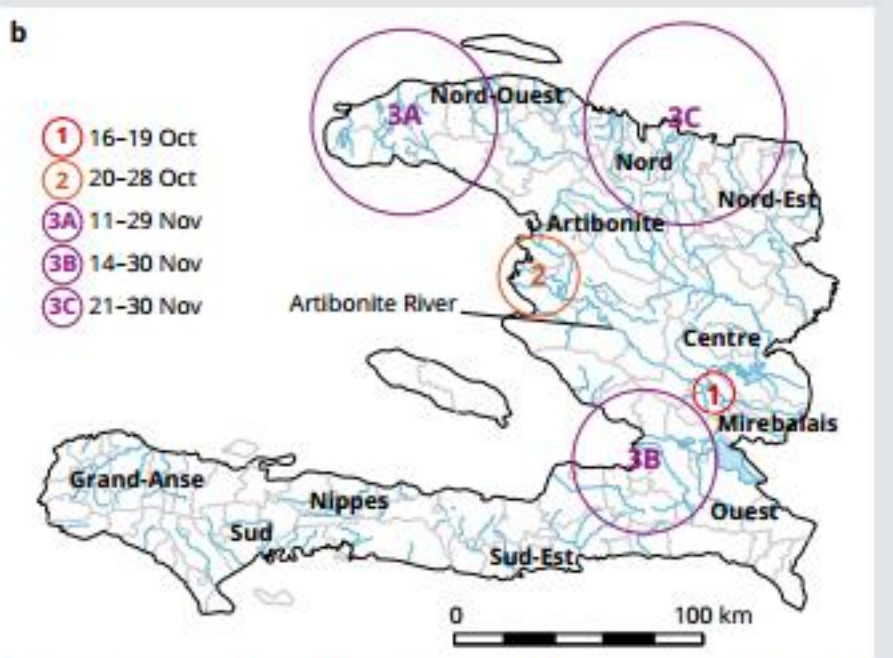
Within weeks of the earthquake, a cholera epidemic broke out in Haiti. This was very surprising to everyone because there had not been any cases there for many decades.

The diagrams on the right show the location of health centres reporting cholera cases on 20 or 21 October 2010, and the progress of the epidemic between its commencement on 16 October through to 30 November, when it had become widespread in Haiti.

a Design a better distribution of the health facilities in order to more effectively limit the spread of disease in times of disaster. Justify your design using the maps supplied, the information provided and your research and knowledge of disease.

b In 2016, another major outbreak of cholera occurred in Haiti. Research the event that caused the outbreak. Explain why there were fears it could be worse than the 2010 outbreak.





(a) Location of health centres reporting cholera cases on 20 October or 21 October 2010 (b) progress of epidemic to 30 November 2010

CHAPTER

08 The immune response

In this chapter, you will learn about how plants and animals resist infection by pathogens and the physiological responses that they have when the barriers fail. You will learn about the innate and adaptive immune responses in mammals, how antibodies are made and long-term immunity is created. You will also learn about vaccination and how it stimulates immunity.

Syllabus subject matter

Topic 2 • Infectious disease and epidemiology

- Explain how host cells recognise self from non-self. **8.1**
- Identify the three lines of defence in vertebrates
  - the innate immune response: skin and mucous membranes (non-specific)
  - inflammatory response and complement system (non-specific)
  - adaptive immune response (specific). **8.1, 8.2**
- Describe the inflammatory response, including the role of
  - prostaglandins and vasodilation
  - neutrophils and macrophages
  - natural killer cells. **8.1**
- Explain the adaptive immune response, including the
  - humoral response (B lymphocytes, antibodies)
  - cell-mediated response (T lymphocytes)
  - role of memory cells. **8.2**
- Compare active and passive immunity, both naturally acquired and artificially acquired. **8.3**
- Interpret long-term immune response data. **8.2, 8.3, Data analysis***
- Describe the innate immune responses in plants, including
  - physical defence strategies: barriers and leaf structures
  - chemical defence strategies: plant defensins and production of toxins. **8.1**
- Interpret data from an experiment investigating the effect of an antimicrobial agent on the growth of a microorganism. **8.3, Practical investigation 5**

*The greyed-out dot point is also addressed in Chapter 9.

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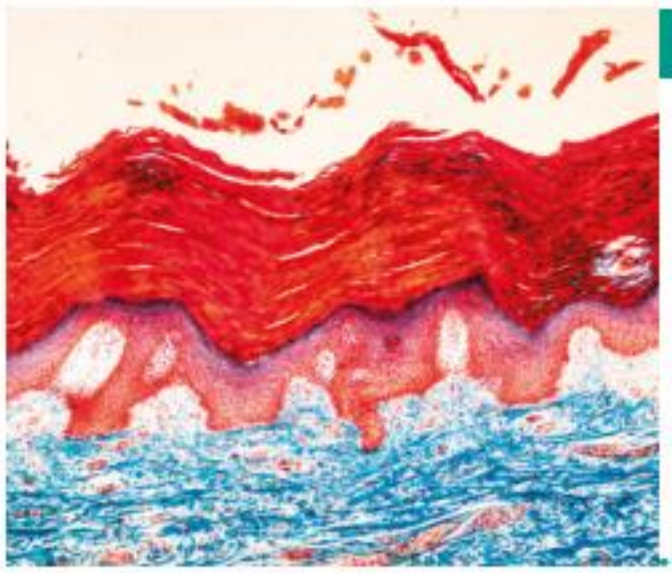
8.1 Innate immunity



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand the role of antigens in immune responses
- know how animals and plants use physical and chemical barriers to resist the entry of pathogens
- be able to describe the steps in the innate immune response in animals and plants
- be able to name and describe the roles of the cells involved in the innate response
- recognise the importance of the major histocompatibility complex in identifying self and non-self.

Antigens are unique molecules, or parts of molecules, that can elicit an immune response, and so play a crucial role in immunity. The mechanisms by which organisms defend against pathogens and identify non-self antigens are multiphase and, in many instances, quite complex. These defence mechanisms include barriers that help prevent infection and immune responses to the pathogens that breach these barriers.

The defence mechanisms of plants and most invertebrates seem to be confined to innate immunity. Innate immune responses are non-specific and do not create immunological memory, so the response to subsequent infections is the same as the first response. Vertebrates have innate responses, like plants and invertebrates, but also have an adaptive, or specific, immune response, which is modified on each exposure to the pathogen, and is highly specific to the particular pathogen.

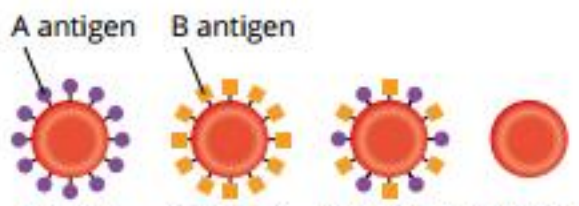


FIGURE 8.1.1 The A and B blood group antigens are carbohydrate molecules attached to proteins and lipids in the cell membrane and displayed on its surface.

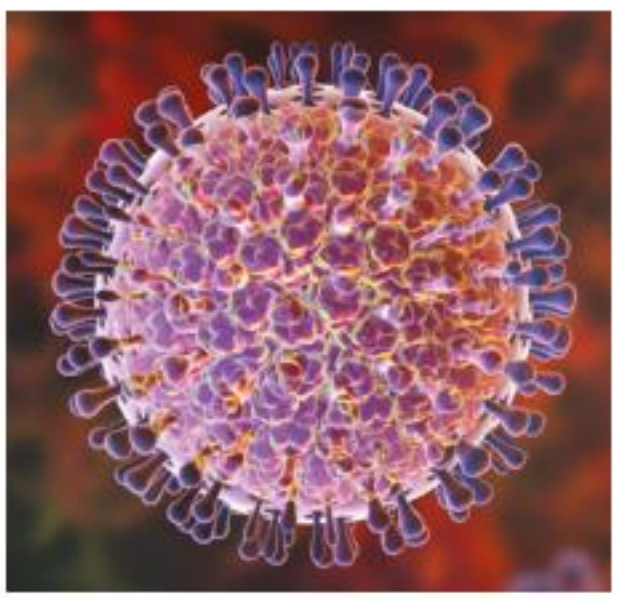


FIGURE 8.1.2 An artist's impression of rotavirus, a virus that is a common cause of gastroenteritis and diarrhoea in infants. Proteins on the surface of the virus act as antigens, which are recognised by the body's immune cells.

ANTIGENS

Antigens are substances that, upon entering the body, can evoke an immune response. Antigens can be classified as self or non-self, and an organism's cells, especially immune system cells, can usually differentiate between self and non-self antigens and respond accordingly. Any antigen that elicits an immune response is more properly known as an **immunogen**. However, in the context of an immune response it is still common to simply refer to them as antigens.

Antigens are expressed or presented on the surface of the cell membrane of cells, where they act as recognition sites for the immune system. However, not all antigens are attached to a cell; for example, some antigens, such as toxins released by bacteria, circulate freely in body fluids. The immune system can normally distinguish antigens that are expressed by its own cells from those that are not and respond accordingly.

An example of antigens is the cell membrane complex carbohydrates of the human ABO blood group. It is the structure of the cell membrane carbohydrate that makes the A antigen different from the B antigen. The presence or absence of A and B antigens on the surface of red blood cells determines whether your blood group is A, B or AB. Group O blood has neither A nor B antigens on the surface of red blood cells. Figure 8.1.1 shows the four major blood groups. Proteins on the surfaces of virus particles or cells also act as antigens, as can be seen in Figure 8.1.2.

Antigens normally present on cells of the body are self-antigens and should not elicit an immune response. The lack of response to self-antigens is called tolerance, or **self-tolerance**. If self-tolerance breaks down and the immune system responds to self-antigens, it results in autoimmune diseases.

Some substances rarely trigger an immune response. For example, earrings are often made of gold because gold does not normally elicit an immune response.

Other substances almost always stimulate an immune response. These include poison ivy (a plant) resin, the venom from insect bites, transplanted tissues and pathogen-associated molecules. There are also some substances, such as various pollens and antibiotics, that sometimes cause a response. The response in this case is an over-reaction to antigens that are of no danger to the body. These antigens are called **allergens**, and the reaction to them is an allergic reaction.

BARRIERS TO INFECTION

Organisms have a number of first-line defences (or barriers) that provide innate resistance against pathogens, including:

- physical barriers, such as skin or bark
- chemical barriers, such as the lysozyme enzymes in saliva and other body secretions
- microbiological barriers, namely microflora.

Plant barriers to infection

Like vertebrates, plants have natural barriers to invasion by pathogens, but, unlike vertebrates, they don't have an adaptive immune system. Plants also lack mobile immune cells that can travel to the site of infection, so every plant cell has to respond to pathogens and use molecules to signal other cells during infections, resulting in non-specific responses. Plants have both physical and chemical barriers as their first lines of defence.

Physical barriers

Plants have a large variety of physical barriers to prevent pathogens from entering their tissues. Physical barriers include:

- cell walls that provide strength and flexibility
- dense cuticles on the outside layers of epidermal tissues, which are made of cutin and other waxes. The thicker the cuticle, the harder it is for pathogens to enter the cells
- thick layers of bark that also prevent pathogen entry
- lignified cell walls. Lignin is the main constituent of wood. It is dense and impervious to water. Lignin is difficult for insects to chew through and if undamaged is impenetrable to pathogens
- the ability to close stomata to deny pathogens entry. Stomata (singular: stoma) are the pores through which gas exchange takes place. Such openings are an ideal entry point for pathogens so the ability to close them when under attack is beneficial. A stoma is shown in Figure 8.1.3
- vertical positioning of leaves, which means water is unable to collect on the surface of leaves. This helps limit infection by pathogens that are reliant on water for motility.

FIGURE 8.1.3 A scanning electron micrograph of a single stomata on the surface of the leaf of a tomato plant. Stomata can close to prevent bacteria (pink, rod-shaped structures) entering and infecting the plant.

Chemical barriers

Plants possess specific genes called resistance genes (R). Resistance genes code for proteins (R proteins), which switch on a plant's defences when it recognises a specific molecule produced by the pathogen. The specific pathogen molecules are generally proteins, known as AVr or avirulence proteins, and are coded for by AVr genes in the pathogen. They are avirulence genes because the presence of their protein product stimulates such a rapid and overwhelming response by the infected plant tissues that the pathogen is rapidly destroyed, and disease does not eventuate.

A plant can mount an immune response if a pathogen enters the plant's tissue, either by responding to AVr proteins or through pathogen-recognition pathways. However, in many cases, immune responses are not required because the initial barriers to infection have held.

When the physical barriers are breached, plants make a range of toxic chemicals that help defend against infection. Many of these chemicals are usually present in the plant tissues, although their levels may increase when the plant is attacked by a pathogen.

Toxic chemicals produced by plants in response to pathogens include saponins, terpenes, phenolics, alkaloids and cyanogenic glycosides.

Saponins have soap-like properties, and like soap, they break down lipids, and so disrupt the cell membranes of pathogens.

Terpenes make up many of the essential oils found in plants. Pyrethrins are terpenes that are used in insecticides. Phytoectysones are terpenes that mimic the hormones involved in insect moulting and cause disruption to larval development, increasing mortality, and thus reduce infestation.

Phenolics include flavonoids, tannins and phytoalexins. Phytoalexins and many flavonoids have antibiotic properties. They disrupt cellular metabolism in the pathogen. Phytoalexins act against bacteria but are particularly effective against fungi where they can disrupt the cell membrane. Phytoalexins are lipophilic and can cross cell membranes and enter the pathogen cells. Inside the cells they inhibit the activity of essential enzymes, causing cell death. Tannins are water-soluble chemicals that are highly toxic to plant pathogens. Tannins are stored in vacuoles until required. They bind to salivary proteins and digestive enzymes such as trypsin in multicellular pathogens. This can result in the death of the pathogen through inadequate energy intake. Tannins are also effective against bacteria where they attach to iron atoms and deny them to the bacteria. Iron is an essential material in bacterial metabolism. Tannins also reduce the ability of bacteria to adhere to host cells which reduces their ability to cause disease. The effects of tannins on bacterial growth are shown in Table 8.1.1. Tannins are effective against even antibiotic-resistant bacterial strains.

Alkaloids are toxic to many organisms, ranging from bacteria to humans. Many alkaloids are highly toxic to fungi, bacteria and insects. Their toxicity is usually dose dependent. Caffeine, nicotine, morphine, capsaicin and atropine are all alkaloids. Some alkaloids are used in low doses as drugs, such as atropine, which is used as a cardiac stimulant in minute doses but is lethal in large doses.

Cyanogenic glycosides are compounds that break down to form hydrogen cyanide (HCN). Hydrogen cyanide is extremely toxic to all eukaryotic cells because it disrupts ATP production in the mitochondria by blocking the electron transfer chain. This quickly leads to cell death.

TABLE 8.1.1 Zone of inhibition of tannins extracted from green tea on growth of selected bacteria. An antibiotic was used as a control. Paper discs were soaked in tannin extract or antibiotic

Mass (mg) tannin per disc	Zone of inhibition (mm)				
	Vancomycin-resistant *Staphylococcus aureus*	Methicillin-resistant *Staphylococcus aureus*	*Bacillus coagulans*	*Shigella flexneri*	*Listeria monocytogenes*
2	0.2–1.5	<0.1	2.0–4.5	0.2–1.5	0.2–1.5
3	2.0–4.5	0.2–1.5	2.0–4.5	0.2–1.5	0.2–1.5
4	2.0–4.5	0.2–1.5	2.0–4.5	2.0–4.5	2.0–4.5
5	2.0–4.5	2.0–4.5	5.0–7.0	2.0–4.5	2.0–4.5
Antibiotic control	5.0–7.0	5.0–7.0	>8	>8	2.0–4.5

If all previous defences fail to prevent pathogens from infecting the plant, cell-mediated defences can involve self-destruction of infected or damaged cells, which helps to limit a pathogen's access to nutrients and, in turn, limit the spread of the pathogen to the rest of the plant. Many plants also produce a hypersensitive response when invaded by parasites, such as nematode larvae or bacteria. This response involves the rapid breakdown of cells around the parasite and the release of toxic substances. This may kill the invader, but it also signals surrounding tissues to increase their levels of resistance to infection.

Animal barriers to infection

Animal barriers to infection are generally different from plant barriers but are just as effective. Unbroken skin and body secretions are among the most important strategies used by animals to prevent pathogens entering the body. These are the first line of defence against disease. Figure 8.1.4 shows the major physical and chemical barriers to infection.

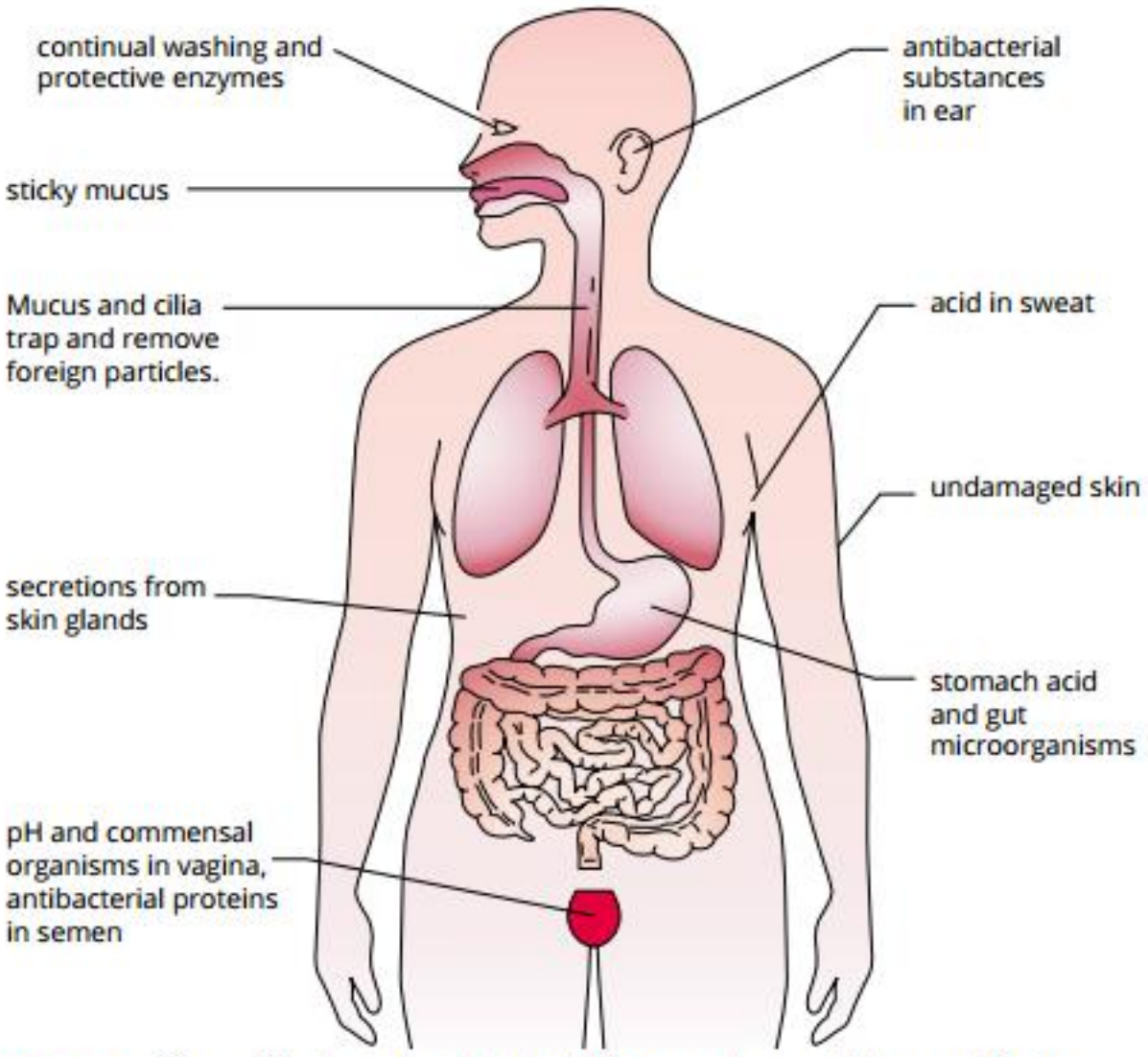

continual washing and protective enzymes
antibacterial substances in ear
sticky mucus
Mucus and cilia trap and remove foreign particles.
acid in sweat
undamaged skin
secretions from skin glands
stomach acid and gut microorganisms
pH and commensal organisms in vagina, antibacterial proteins in semen

FIGURE 8.1.4. Some of the physical and chemical defence mechanisms that prevent foreign organisms from gaining access to the human body.

Physical barriers

In animals, epithelial cells create a physical barrier that prevents pathogens from entering the organism. Epithelial cells line the skin, as well as the respiratory, gastrointestinal and urogenital tracts. They are joined tightly by specialised membrane proteins to form a continuous barrier against pathogens.

In addition to toughened (keratinised) unbroken skin, physical barriers to pathogens in animals include mucus-secreting membranes. Invading organisms are trapped in mucus and membranes lined with cilia sweep foreign bodies away (e.g. those that line the airways of mammals). These can be seen in Figure 8.1.5.

Chemical barriers

External chemical barriers in vertebrates include lysozyme enzymes and toxic metabolites, such as lactic acid and fatty acids, which are found in secretions such as tears, sweat and saliva. Here, they have protective functions and provide a generalised defence; for example, by destroying bacterial cell walls.

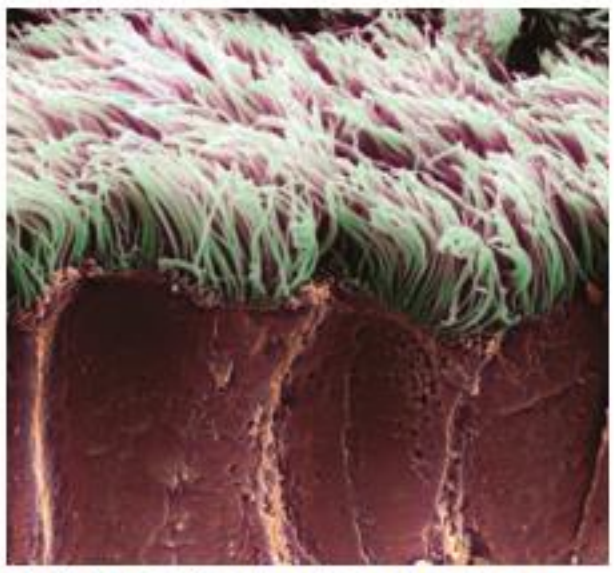


FIGURE 8.1.5 A coloured scanning electron micrograph of the mucous membrane (or bronchial epithelium) that lines the major airways of the lung. Mucus traps potential pathogens and foreign particles, and the rhythmic movement of hair-like cilia moves bacteria and other particles away from the lung and towards the throat.

Other chemical barriers include stomach acid and digestive enzymes, which are primarily involved in the digestion of food, but also kill many pathogens. The fluid in the lungs contains proteins that act as surfactants. Surfactants are 'detergent-like' substances found in lung secretions that lower the surface tension of lung fluids and prevent the alveoli from collapsing on exhalation. They are also antimicrobial because they coat pathogens, making it easier for the pathogens to be eliminated by **macrophages**. In female mammals, the lining of the vagina is coated in acidic secretions that serve several functions, including defence against pathogens.

Microbiological barriers

In healthy people, non-pathogenic bacteria, referred to as normal flora, are found on the skin, and in the mouth, nose, throat, lower part of the gastrointestinal tract and the urogenital tract. The presence of normal flora prevents the growth and colonisation of other bacteria because normal flora competes with other bacteria for space and resources, and produces chemicals that lower the pH of the micro-environment.

If you take a course of antibiotics, you can disrupt the normal flora of your intestine and reproductive tract, because the antibiotics do not discriminate between beneficial normal flora and harmful bacteria. This can disturb the normal function and predispose you to various infections until the levels of normal flora return to their pre-treatment values. For example, the use of antibiotics can allow the growth of yeast infections. Yeasts are fungi and are not affected by most antibiotics. The yeast *Candida albicans* is generally present in most humans in the mouth and gut and in the vagina of females. Disruption to the normal bacterial suite, which keeps the size of the yeast colony under control, can result in the disease—thrush.

In healthy people, competition between all of the natural flora keeps bacterial numbers in balance. However, in people with weakened immune systems, normal flora can sometimes grow unchecked and cause disease.

INNATE IMMUNE RESPONSES

If pathogens manage to breach the first line of defence, they are immediately met by attacking cells and molecules. This line of defence is known as the **innate immune response**.

The innate immune response exists in all organisms, and its persistence over millions of years of evolution indicates its fundamental importance. Even if the innate immune response cannot eliminate a pathogen, it is still critical for keeping infections under control until the adaptive immune response (see Module 8.2) starts working, which can take up to several days.

Innate immune responses in vertebrates:

- are non-specific—they do not target a specific antigen
- are rapid—they occur within hours
- are present in all animals
- are fixed responses—they do not adapt
- do not lead to an immunological memory of the pathogen that caused the infection.

Plant responses to infection

The main response of plants to infection is a chemical response. Plants make a variety of chemicals that disrupt pathogen reproduction. Some of these chemicals are present in plant tissues all of the time (the chemical barrier), whereas others are only produced during infection or potential infection. The plant response is triggered when plant cells recognise certain molecules, such as certain lipopolysaccharides, or other common cell wall components, which form the cell walls of pathogens.

These molecules are called **microbe-associated molecular patterns (MAMPs)** and are recognised by **pattern recognition receptors (PRRs)**. Plants use hormone-like chemicals, such as jasmonic acid and salicylic acid, to activate their responses in the recognition pathways. The plant response pathway is summarised in Figure 8.1.6.

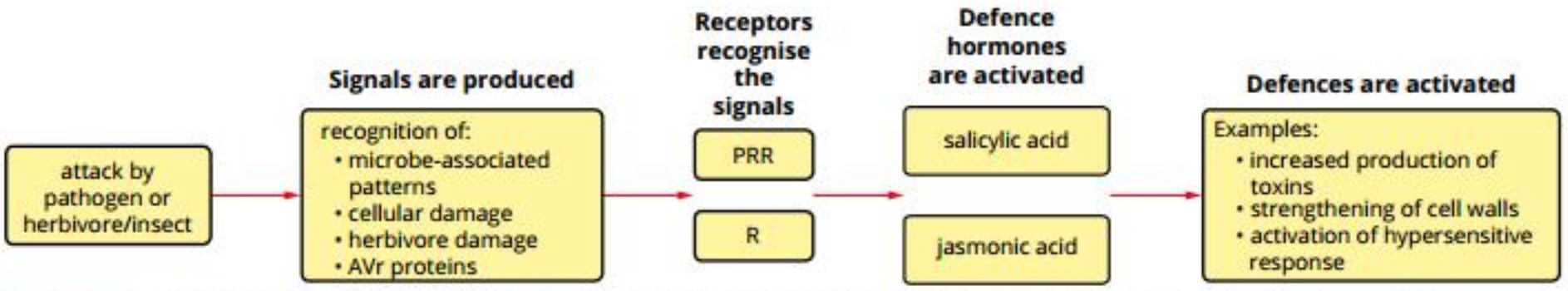


FIGURE 8.1.6 Plants recognise attack by pathogens or plant-eating organisms and mount defences that strengthen natural barriers and increase production of toxic and signalling chemicals.

Plants produce a range of proteins and enzymes that provide defence against pathogens. Because it requires a lot of energy to produce these substances, most plants only make them when under actual attack by a pathogen.

Among the proteins produced by plant tissues are:

- **defensins**, which are small proteins that act against digestive enzymes and are also thought to act against microbes by disrupting the cell membrane. Many of these proteins are rich in the amino acid, cysteine. Bread wheat, shown in Figure 8.1.7, can produce defensins
- **protease inhibitors**, which inhibit enzymes such as trypsin, an important digestive enzyme
- **digestive enzyme inhibitors**, which are proteins that block normal digestion. They include enzymes, lectins, which block the digestion of starch by insects, and ricin, which is so toxic that 0.2 mg is enough to kill an adult human
- **hydrolytic enzymes**, which break down cell walls. Chitinases break down chitin, the main constituent of fungal cell walls and insect exoskeletons. Glucanases break the chemical bonds between the molecules that form glucans, which comprise the cell walls of members of the oomycetes. Lysozymes are enzymes that can digest bacterial cell walls.

In many plants, recognition of a pathogen may also activate enzymes that strengthen the cell wall as a barrier to infection and cause surrounding cells to thicken their cell walls. Many plants also undergo cell-mediated responses when all other pathways have failed. These responses can result in the self-destruction of the infected tissues, the hypersensitive response, in an attempt to wall off the pathogen and so protect the rest of the plant.

FIGURE 8.1.7 Bread wheat (*Triticum aestivum*) contains small cysteine-rich proteins that act as plant defensins to inhibit the growth of bacteria and fungi.

Animal responses to infection

Like plants, animals can recognise and respond to pathogens through the identification of **pathogen-associated molecular patterns (PAMPs)**. In vertebrates, this response includes a large range of responses and involves many specialised cells, especially white blood cells (or **leukocytes**).

The vertebrate response to pathogens is probably the most complex of all organisms. It includes both innate and adaptive components. The **adaptive immune response** provides long-term immunity to pathogens through the creation of immunological memory. The adaptive response is modified on each subsequent infection and becomes more effective each time. Many different types of cells are involved in both the innate and adaptive responses in vertebrates. In this section, the focus is on the innate response of mammals, especially humans.

Defensive molecules

Complement proteins and cytokines are defensive molecules involved in both the innate and adaptive immune responses.

The **complement system** comprises more than 30 **complement proteins** that circulate in the blood and help kill foreign cells. They exist in body fluids in an inactive form and are activated as part of the non-specific immune response to certain antigens and carbohydrates on the surfaces of some bacteria and parasites.

When the first protein is activated, it triggers a cascade of reactions, each protein activating the next in the cascade.

The signals that activate the complement cascade include **antigen–antibody complexes** of the immune response and carbohydrates on the 'foreign' surfaces of some bacteria and parasites. Activated complement proteins have a number of important actions, such as:

- attracting and enhancing the activity of phagocytic cells, which assists them to recognise and engulf foreign material
- forming attack complexes to destroy bacteria directly by punching holes in bacterial membranes and causing the bacterial cell to breakdown (lyse) and release their contents, which attracts phagocytes to the site of infection. This is shown in Figure 8.1.8
- contributing to inflammation by increasing local permeability of capillaries and attracting phagocytes.

The complement system is a part of the innate immune response. However, it can be recruited by the adaptive response through the binding of antibody–antigen complexes to certain complement proteins.

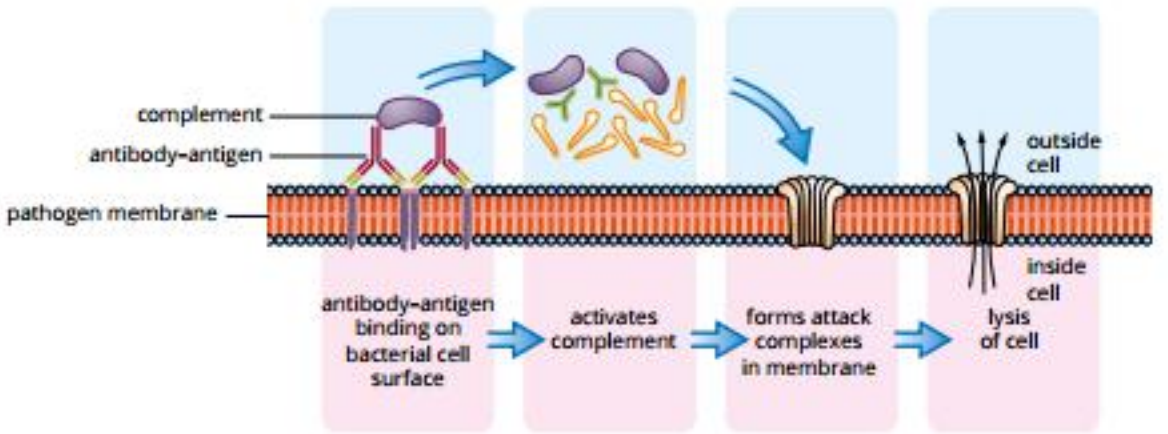


FIGURE 8.1.8 Complement proteins activated by antigen–antibody complexes are also involved in specific (adaptive) immune responses.

Cytokines are small signalling molecules used by the immune system. They coordinate many aspects of immune responses. Cytokines can be peptides, proteins or glycoproteins, and are released by body cells in response to cell damage or the presence of pathogens. There are many different cytokines, and they trigger a variety of responses, both non-specific and specific. For example, cytokines can promote the rapid increase of **lymphocytes**, induce inflammation and fever, activate macrophages and regulate cells in the adaptive immune response. Interferons and chemokines are two examples of cytokines, and they each have different functions.

Interferons are a class of cytokines that are produced by, and act on, a host cell infected by a virus. Interferons stimulate the infected cells to produce enzymes that break down viral RNA and proteins that block translation. This limits viral replication and release from the cell. Interferons also attract natural killer cells, which release cytotoxic peptides to kill the virus-infected cell. Interferons are non-specific and act against any virus. However, viruses vary widely in their susceptibility to interferons. Many viruses can evade interferon-induced defences and the more virulent viruses may be able to inhibit the production of interferon.

Interferons are an important defence against viruses but play a smaller role in combating bacterial and parasitic infections. Interferons also regulate the immune system in a number of ways, such as enhancing T lymphocyte activity.

Chemokines are a group of cytokines that act as chemical attractants (or chemo-attractants). Chemokines are important for attracting leukocytes to sites of infection and inflammation.

Cells of the innate immune response

Leukocytes are immune cells that are present in blood and other tissues. Leukocytes have pattern recognition molecules, also known as toll-like receptors (TLRs) on their surface, which are able to recognise PAMPs. Different TLRs recognise different PAMPs. For example, TLR-2 recognises the lipoproteins and peptidoglycan of gram-positive bacteria, TLR-4 recognises lipopolysaccharides of gram-negative bacteria, TLR-5 recognises bacterial flagellin, and TLR-7 and TLR-8 recognise single-stranded viral RNA.

The cells of the innate immune system of mammals include:

- macrophages
- neutrophils
- monocytes
- dendritic cells
- natural killer cells.

Natural killer cells identify and kill somatic (body) cells that have been infected by viruses. They do this by producing chemicals called **perforins**, which perforate the cell membrane. This causes cell contents to leak from the cell and results in **phagocytes** engulfing and digesting the dying cell. Natural killer cells may also introduce **granzymes** into the cell. Granzymes are enzymes that cause the cell to undergo **apoptosis** (programmed cell death). The granzymes trigger the activation of other enzymes which either start breaking down the cell and trigger the activation of more enzymes along with attracting phagocytes. Ultimately, the cell is completely destroyed, and the debris is digested by the phagocytes.

Macrophages, neutrophils, monocytes and dendritic cells are all phagocytes. Phagocytes engulf pathogens and the remains of dead cells through the process of phagocytosis. Neutrophils are the most abundant of all white blood cells and are usually the first leukocytes to arrive at the site of infection. Monocytes are only found in the blood. Once they leave the blood, they mature into macrophages or dendritic cells. Like neutrophils, macrophages and dendritic cells phagocytose pathogens and toxins and destroy them. However, macrophages and dendritic cells can also process the antigens from the pathogen or toxin and use these to stimulate the adaptive immune response discussed in the next module. You can see examples of phagocytes in Figures 8.1.9 and 8.1.10.

TLRs on the phagocyte interact with a microbe's PAMPs to cause a series of changes inside the cell and activate the phagocyte. Once activated, the phagocyte engulfs the microbe, with the cell membrane forming a vacuole called a **phagosome** around it. Then a lysosome containing digestive enzymes fuses with the phagosome, forming a **phagolysosome**, which breaks down the foreign material. The fragments can then be expelled from the cell by exocytosis. Exocytosis is the release of substances enclosed within a vesicle to the outside of a cell. It occurs by fusion of the vesicle with the cell membrane.

Table 8.1.2 on page 322 shows a more detailed list of the leukocytes involved in the innate immune response and indicates whether they are involved in phagocytosis, antigen presentation or the release of cytokines that promote inflammation. You will learn more about cytokines later in this module.

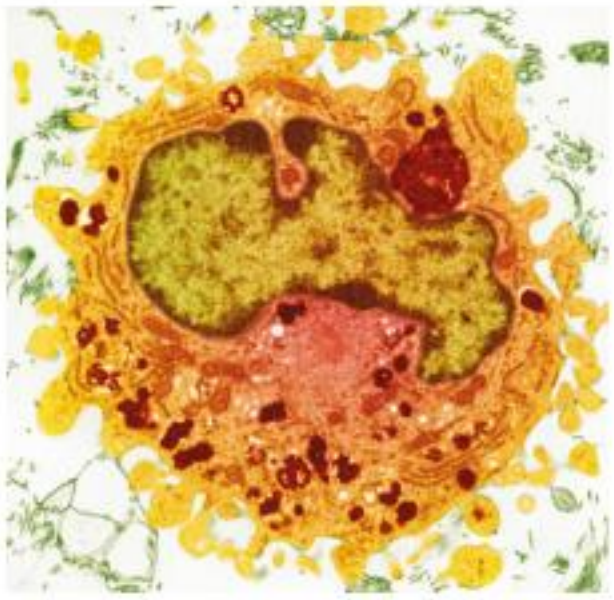


FIGURE 8.1.9 A coloured transmission electron micrograph of a macrophage. Macrophages are cells of the innate immune response in vertebrates that recognise and engulf foreign material.

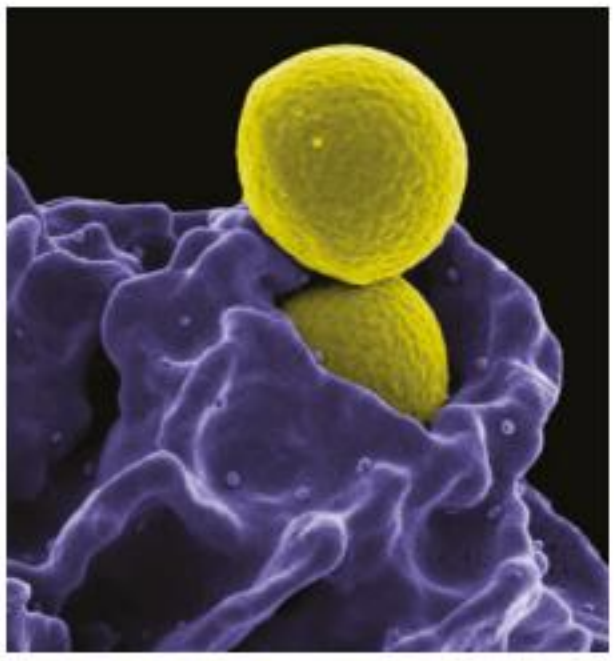
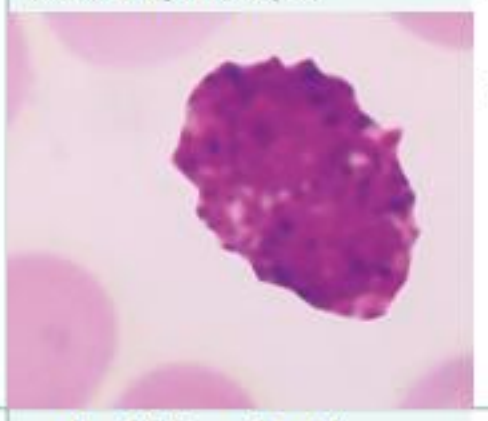
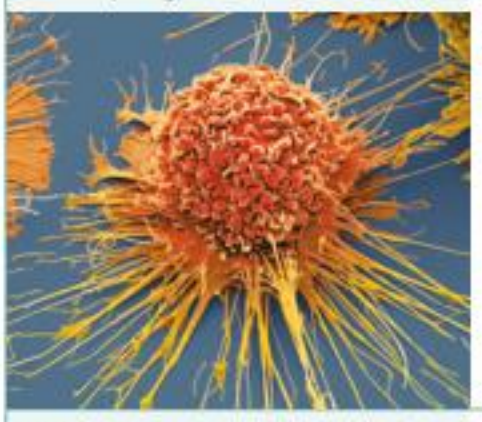
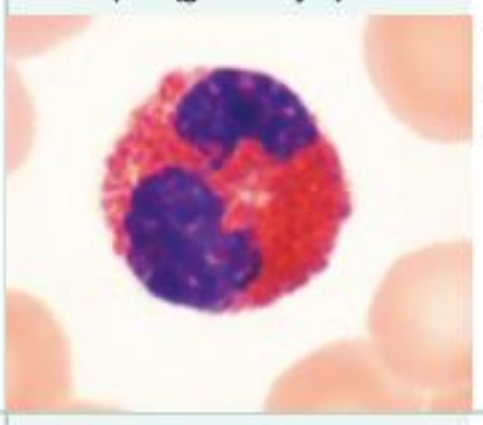
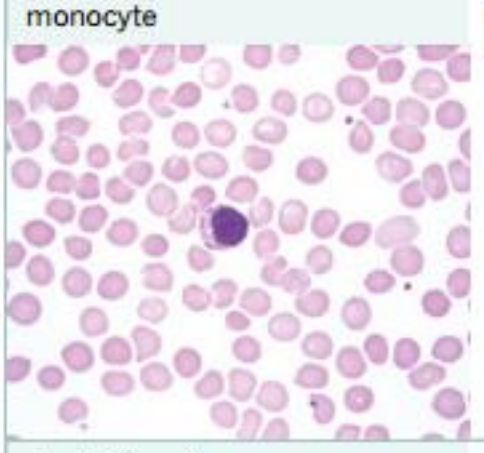
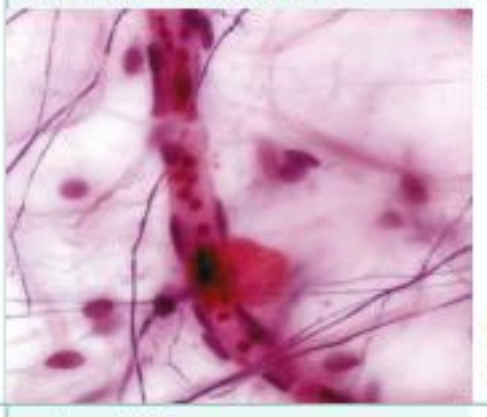
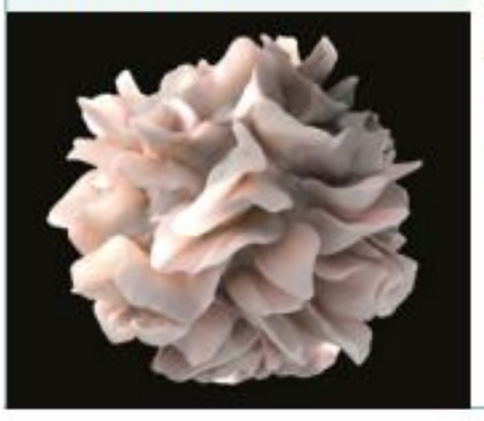
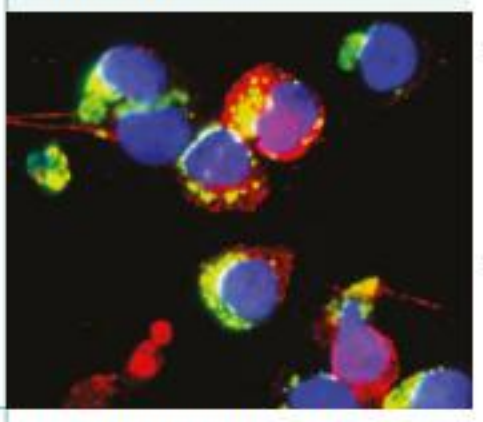


FIGURE 8.1.10 Phagocytes are a key part of the innate immune response. This neutrophil (purple) is engulfing *Staphylococcus aureus* bacteria (yellow), which it then phagocytoses.

TABLE 8.1.2 A more detailed list of the leukocytes involved in the innate immune response and their functions

Cell type	Function	Cell type	Function
neutrophil (granulocyte) 	• phagocytosis • releases antimicrobial compounds, such as defensins and hydrogen peroxide, which disrupt bacterial and fungal membranes • releases cytokines that attract other immune cells and cause inflammation	basophil (granulocyte) 	• releases histamine, which contributes to inflammation and therefore blood vessel dilation • has a limited role in phagocytosis
macrophage 	• phagocytosis • antigen presentation • releases cytokines	eosinophil (granulocyte) 	• antigen presentation • releases cytokines and cytotoxic chemicals • has a limited role in phagocytosis • found in large numbers in parasitic infections
monocyte 	• phagocytosis • mature into macrophages or dendritic cells after leaving the bloodstream	mast cell (granulocyte) 	• plays a key role in inflammation, and therefore blood vessel dilation, by releasing histamines • has a limited role in phagocytosis Mast cells stained red/pink with haematoxylin and eosin stain
dendritic cell 	• phagocytosis • antigen presentation • has many grooves that increase surface area and permit contact with a large number of other cells	natural killer 	• recognises virus-infected and cancerous cells • releases cytotoxic chemicals from granules, such as perforin, which makes holes in cell membranes, triggering apoptosis and cell death of virus-infected cells and abnormal cells • releases cytokines to attract and activate cells of the adaptive immune system An immunofluorescent light micrograph of natural killer cells: cytotoxin granules (green), nuclei (blue), cytoplasm (red)

Inflammatory response

Inflammation is the accumulation of fluid, plasma proteins and leukocytes that occurs when tissue is damaged or infected. Inflammation results in heat, pain, swelling, redness and loss of function at the site of infection and may involve body-wide effects such as fever.

Inflammation is often triggered by substances released from damaged cells, by **histamines** released by stimulated **mast cells** and by **platelets**. Mast cells are stationary granular cells located in the walls of blood vessels, gut and lungs. Platelets are membrane-bound fragments of huge white blood cells (**megakaryocytes**) located in the bone marrow. Platelets are about half the size of red blood cells and have no nucleus or DNA. Megakaryocytes 'pinch off' thousands of platelets during their life and release them into circulation where they remain for about 10 days. They are important in wound healing and blood clotting.

The interaction between leukocytes (especially phagocytes) and pathogens may also trigger the inflammatory response that results from the production, activation or release of peptides and proteins such as complement proteins and cytokines.

Acute inflammation involves phagocytes and occurs soon after infection as part of the innate immune response. Inflammation can also involve lymphocytes and occur later as part of the adaptive immune response.

A number of steps are involved in the initiation of an inflammatory response to infection. These steps are shown in Figure 8.1.11.

1 Bacteria and other pathogens breach the barriers of the first line of defence; for example, through an open cut or wound in the skin.

2 Injured cells release cytokines (chemokines) that attract neutrophils, and mast cells release histamine, which increases **vasodilation** (blood vessel dilation) and permeability. The dilated, more permeable blood vessels, allow leukocytes and fluid containing peptides and proteins such as complement proteins to enter the infected tissue. Platelets release clotting factors at the site of the wound.

3 Neutrophils migrate towards the cytokines and are activated, causing the neutrophils to recruit macrophages and secrete factors, such as defensins and hydrogen peroxide, that degrade and kill pathogens; for example, by damaging bacterial cell membranes.

4 Macrophages become activated and secrete cytokines and, along with neutrophils, phagocytose pathogens and debris at the site of infection. This may lead to pus, which is fluid containing leukocytes, dead pathogens and cell debris.

5 The inflammatory response continues until the pathogen is eliminated and the wound has healed.

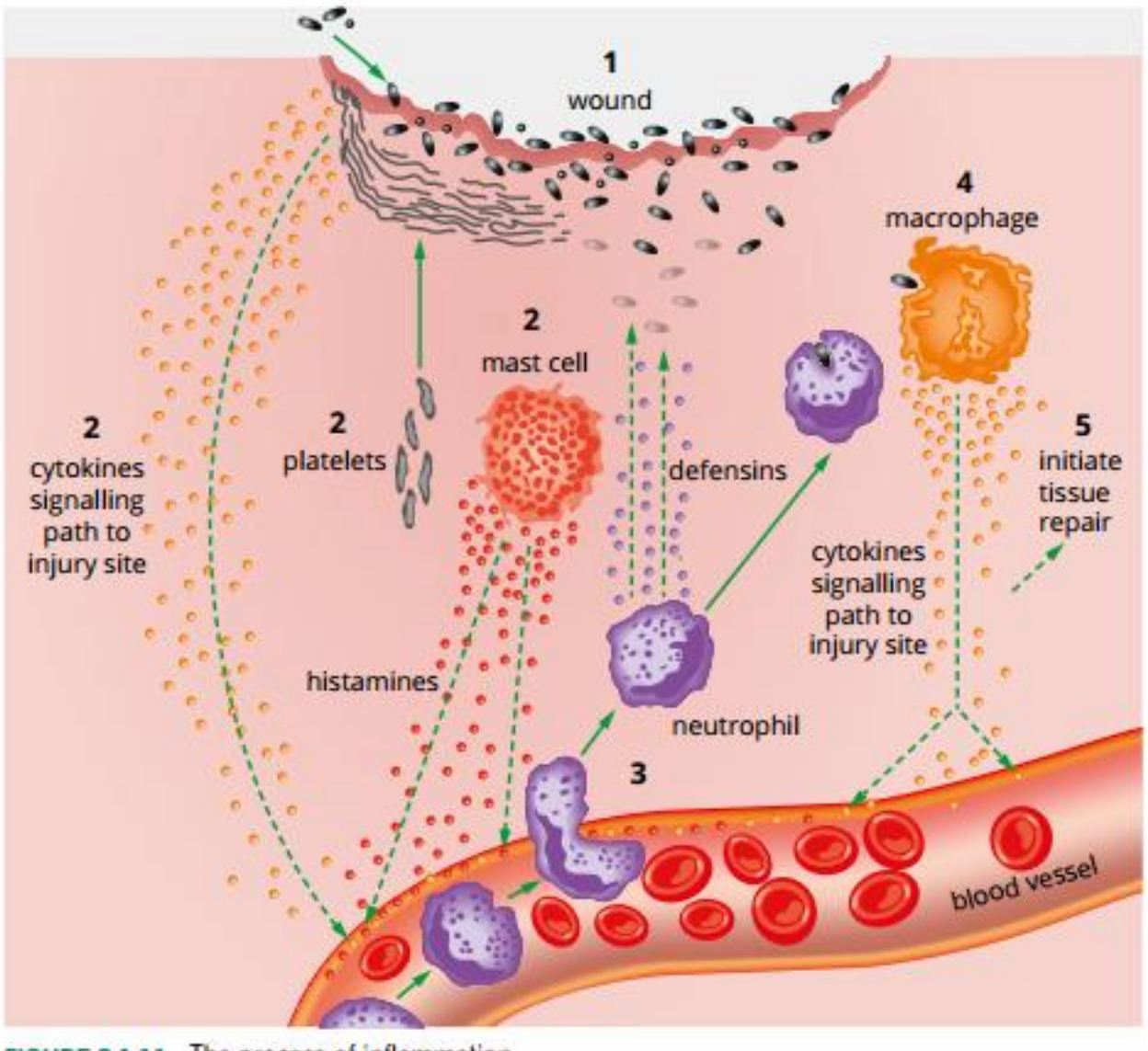


FIGURE 8.1.11 The process of inflammation

Fever

Fever is one common consequence of inflammation. In humans, normal body temperature is about 37°C. Fever occurs when body temperature is above normal. The increase in body temperature results when the regulated body temperature set point in the hypothalamus of the brain is set to a higher level by inflammatory cytokines, including prostaglandins.

Fever slows the replication of bacteria and viruses by shifting the temperature away from their optimal range, and so allows more time for other defences to intervene. Additionally, moderate increases in temperature increase the activity and proliferation of leukocytes, so fever also improves the immune response.

Prostaglandins

Prostaglandins are a group of lipid compounds with hormone-like effects that are involved in many processes. For example, they help to control reactions that cause pain, fever and the healing process and significantly contribute to inflammation. Prostaglandins are involved in stimulating blood vessel dilation (vasodilation). Where pathogens are present and releasing toxins, prostaglandins mediate increased blood flow (through vasodilation) and chemical signals that summon white blood cells. They also stimulate the formation of blood clots if blood vessels are damaged to try to decrease access to tissues for pathogens. As long as pathogens are present and releasing toxins, prostaglandins will continue to be produced and add to the inflammatory process. When the pathogens are finally destroyed a different group of prostaglandins help to resolve the inflammation by reducing vasodilation and breaking up blood clots.

Major histocompatibility complex and antigen presentation

All body cells have **major histocompatibility complex (MHC)** proteins on the surface of their cell membranes. The MHC proteins, also called **human leukocyte antigens** (HLA), are found on the surface of your body's cells, presenting self or non-self antigens to **lymphocytes** (a type of leukocyte primarily involved in the adaptive response; you will learn more about lymphocytes in Module 8.2). MHC proteins are coded for by a gene region, containing over 200 genes, found on chromosome 6.

MHC proteins play a fundamental role in immunity. They are a critical link between the innate and adaptive responses because they are involved in antigen presentation. Two classes of MHC proteins that are important in antigen presentation are MHC class I (MHC-I) and MHC class II (MHC-II). All body cells, except red blood cells, usually have MHC-I markers, however, cells that have been invaded by viruses normally lack MHC-I proteins. The absence of these markers can be recognised by certain cells of the innate immune system, such as natural killer cells, which then initiate the destruction of the infected cell by releasing cytotoxic chemicals, such as perforin and granzyme. In healthy cells the MHC-I markers display self-antigens. The combinations of these antigens are unique to each individual. As their production is genetically controlled close relatives will usually have more self-antigens in common than unrelated individuals.

The two classes differ in structure and function because they are specialised to present different types of antigens, and so elicit different responses.

Some phagocytes, particularly macrophages and dendritic cells, act as **antigen-presenting cells (APCs)**. When APCs phagocytose a pathogen, fragments of digested antigen are linked to MHC-I proteins on the cell membrane, as shown in Figure 8.1.12. The phagocyte then displays, or presents, this pathogen-derived antigen to other cells in the immune system.

In this way, the adaptive immune response is initiated. There are a number of different types of lymphocytes involved in the adaptive response. You will learn more about the cells of the adaptive response in Module 8.2.

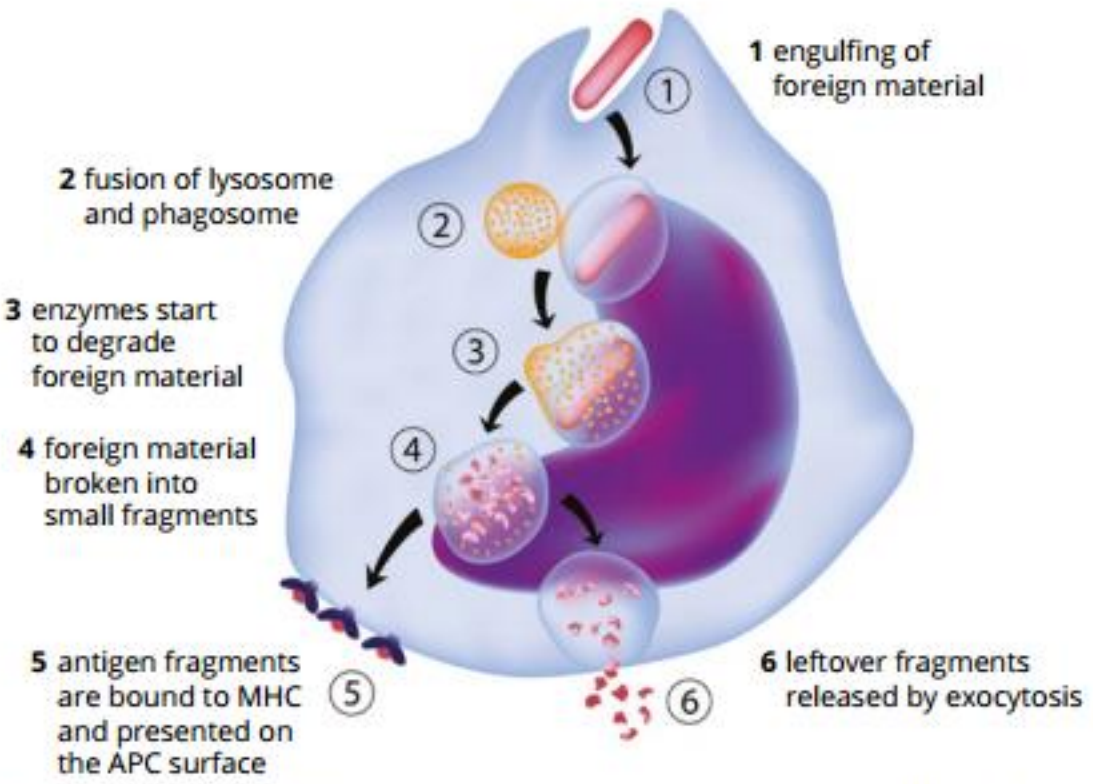

1 engulfing of foreign material
2 fusion of lysosome and phagosome
3 enzymes start to degrade foreign material
4 foreign material broken into small fragments
5 antigen fragments are bound to MHC and presented on the APC surface
6 leftover fragments released by exocytosis

FIGURE 8.1.12 Phagocytosis and antigen presentation in an antigen-presenting cell (APC). APCs communicate with other immune cells by presenting antigens or fragments of antigens on their cell surface.

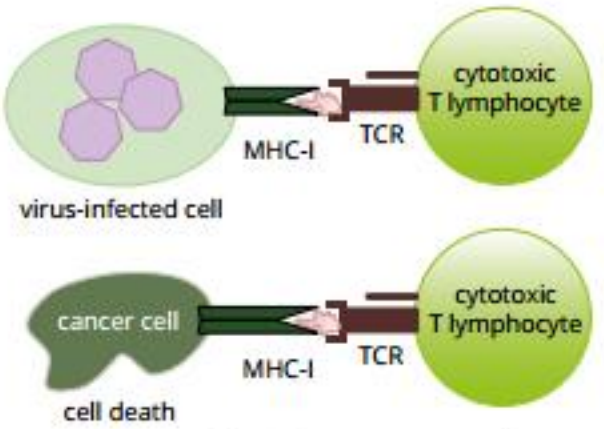


FIGURE 8.1.13 Infected or cancerous cells present antigens on MHC-I to the T cell receptors (TCRs) of cytotoxic T lymphocytes. Cytotoxic T lymphocytes then release cytotoxins that kill the virus-infected cells and cancer cells.

MHC-I proteins also communicate with a type of lymphocyte called a **cytotoxic T lymphocyte (T_C)** about the proteins being produced within each cell, such as non-self viral antigens produced by virus-infected cells. If a T_c lymphocyte reacts to the antigen being presented, it becomes activated and releases toxic peptides (perforin and granzyme) that damage the target cell membrane and induce apoptosis. Figure 8.1.13 shows this process.

MHC-II proteins can be conditionally expressed on all cells, but are most commonly found on the surface of APCs such as dendritic cells, macrophages and B cells, which are cells of the adaptive response. MHC-II presents antigens that originated extracellularly and have been processed after phagocytosis. This presentation of antigens activates the **helper T lymphocytes (T_H)** of the adaptive immune response, linking the innate and adaptive immune responses as the APC and T_H cell interact. Figure 8.1.14 shows this interaction.

Once the antigens are presented, T_H lymphocytes are activated, and secrete cytokines that activate and attract other immune cells to the site of infection, as shown in Figure 8.1.15.

You will learn more about the adaptive immune response and the role of T_H cells in Module 8.2.

FIGURE 8.1.14 A coloured scanning electron micrograph showing the interaction between a macrophage (pink) and a helper T lymphocyte (light green).

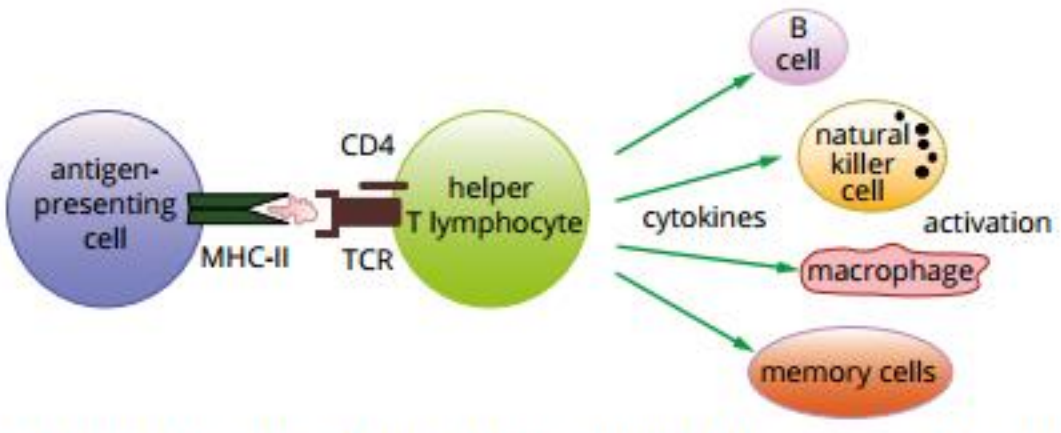


FIGURE 8.1.15 Antigen-presenting cells present antigens on MHC-II to helper T lymphocytes via the T cell receptor (TCR).

8.1 Review

SUMMARY

- Antigens are molecules, or parts of molecules, that are identified by cells of the immune system as self or non-self.
- Antigens are usually composed of one or more polypeptide chains, but may be carbohydrates, nucleic acids or lipids.
- Antigens are found on the surface of cells or free in body fluids.
- Most foreign molecules are identified as non-self antigens and elicit an immune response.
- Self-antigens do not usually elicit an immune response.
- Pathogens are a source of non-self antigens.
- Plants have physical barriers against invasion by pathogens; these include thick cuticles.
- Plants have chemical defences, which may be present in their cells all of the time, such as tannins, or which may be produced when the plant is under attack, such as defensins.
- Vertebrates have a range of non-specific defences against pathogens, including intact skin, phagocytes, complement proteins, natural killer cells and a variety of chemicals, such as interferons.
- Interferons are produced by virus-infected cells. Interferons make surrounding cells more resistant to viruses by interfering with viral reproduction.
- Interferons belong to a group of signalling molecules called cytokines.
- Cytokines are the communication molecules of the immune system.
- The complement system causes the lysis of bacteria, which increases the activity of phagocytes and promotes inflammation.
- Natural killer cells are a part of the non-specific immune response; they are active against virally infected cells and cancer cells.
- Phagocytes are leukocytes that engulf and digest pathogenic antigens and then display those antigens attached to MHC-II markers to the cells of the adaptive response.
- Prostaglandins are important mediators of inflammation, increasing inflammation while infection exists and then reducing it once the infection is resolved.
- Inflammation increases blood flow to an area of infection, bringing more leukocytes, complement proteins and platelets to the area.
- Fever, associated with inflammation, inhibits bacterial growth and enhances the activity of immune system cells, especially phagocytes.
- Vertebrates have a group of genes called the major histocompatibility complex (MHC), which code for MHC-I and MHC-II proteins, which are used for displaying antigens and identifying self and non-self.

KEY QUESTIONS

Describe

1 Define 'antigen'.

2 Describe how phagocytes link the innate and adaptive immune responses.

3 Describe how natural killer cells kill virally infected somatic cells.

Apply

4 Draw the process of inflammation that occurs when bacteria enter skin through an open wound. Label key white blood cells and the molecules they produce in response to the pathogen, and number the steps involved in the inflammatory response.

5 Explain how prostaglandins contribute to inflammation.

6 How do cells such as natural killer cells recognise that a body cell has been infected by a pathogen?

7 Compare the physical barriers in plants with those in animals.

8 Explain what is meant by self-tolerance. In your answer, explain why it is necessary.

9 Mild infections such as the common cold are a regular experience of children attending childcare. One symptom of these infections is a mild fever (up to 39°C). The usual treatment given to children is a medication such as paracetamol, which reduces their temperature to normal.

Explain why reducing the body temperature of a patient with a mild fever may prolong the infection.

10 A patient requires a blood transfusion. The only blood available is either group B or group O.

a If the patient is blood group A, explain which blood should be used for the transfusion.

b Explain whether you would have chosen differently if the patient had been group AB.

11 After a course of antibiotics, some doctors advise their patients to consume a probiotic, which contains bacteria. Explain the potential benefits.

12 Explain why a kidney transplant from a sibling (brother or sister) or a parent is more likely to be successful than from a stranger.

Analyse

13 Cranberry juice contains polyphenols shown to have bactericidal properties. A study was undertaken to investigate the effects of cranberry juice concentrate on the ability of bacteria in the mouth to form biofilm colonies on the teeth. The formation of the colonies in humans increases tooth decay and other diseases of the teeth and gums. Six bacterial species (*Streptococcus oralis*, *Actinomyces naeslundii*, *Veillonella parvula*, *Fusobacterium nucleatum*, *Porphyromonas gingivalis*, and *Aggregatibacter actinomycetemcomitans*) known to cause oral problems were tested. The bacteria were exposed to the cranberry extract for 30 seconds and 60 seconds in the test subjects while the controls were not exposed to any known inhibitor of growth. The results of the experiment are shown in the table below.

TABLE 1 Percentage reduction in bacteria biofilm after exposure to cranberry extract.

Bacteria tested	Percentage reduction in viable bacteria in comparison to the control	
	30 seconds exposure	60 seconds exposure
S. oralis	98.9	98.9
A. naeslundii	65.7	45.4
V. parvula	66.7	72.5
F. nucleatum	39.5	75.3
P. gingivalis	35.3	39.3
A. actinomycetemcomitans	5.6	11.5

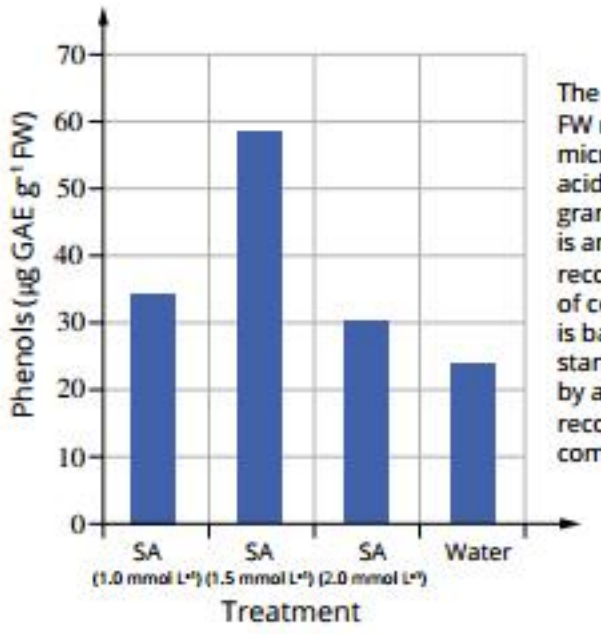
a Against which bacterial species is cranberry extract most effective?

b Against which species is it least effective?

c In which test do the results seem to be anomalous? Justify your answer.

14 It has been suggested that salicylic acid is involved in stimulating the pathway leading to the production of phenols—chemicals that defend plants from attack by pathogens and herbivores.

The graph shows the results after a group of plants were sprayed with salicylic acid (SA) at various concentrations.



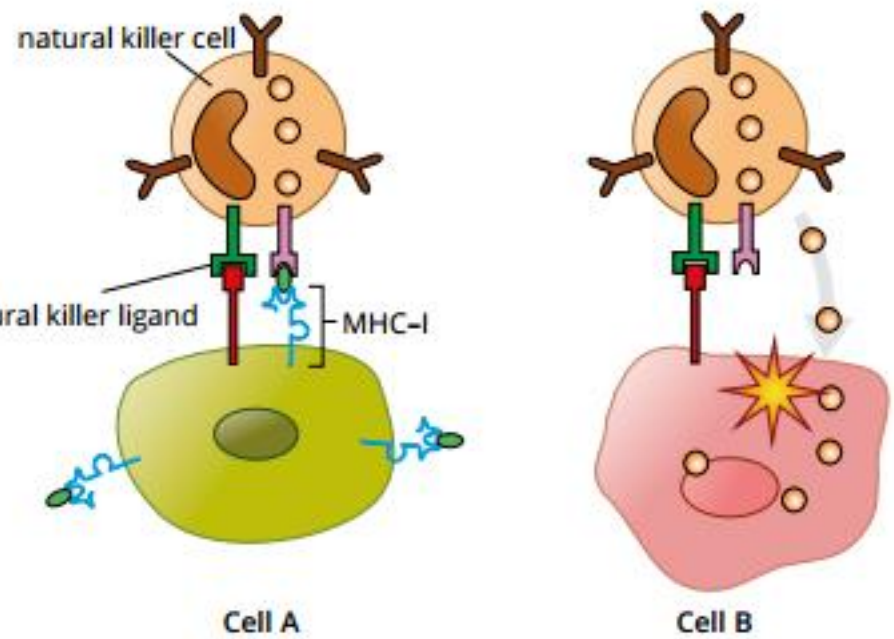
The unit µg GAE g^{-1} FW means micrograms of gallic acid equivalent per gram fresh weight. It is an internationally recognised measure of concentration that is based on a standard table issued by an internationally recognised scientific committee.

a Describe the features of the graph that support the contention that salicylic acid promotes the production of defensive chemicals in plants.

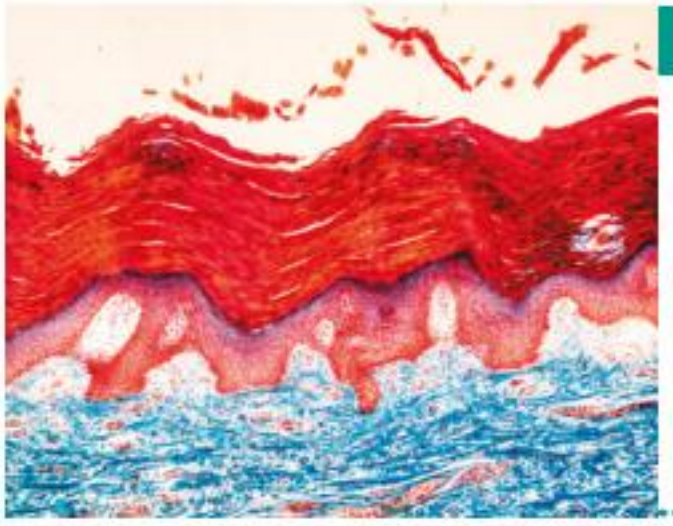
b Identify the concentration of salicylic acid that is most effective and state how you know this.

15 The diagram shows a natural killer cell interacting with two different cells that it has encountered in the body.

Examine the diagram carefully. Explain, using evidence, which cell is being killed and how the natural killer cell identified that the other cell was not a threat.



8.2 Adaptive immunity



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- know the names and roles of the various cells involved in immune responses
- understand the difference between innate and adaptive immunity
- understand the role of antibodies in the adaptive response
- understand how humoral and cell-mediated immune responses provide protection against pathogens
- understand how the immune system identifies which cells to target during immune responses.

FIGURE 8.2.1 Each B lymphocyte produces antibodies for a specific antigen.

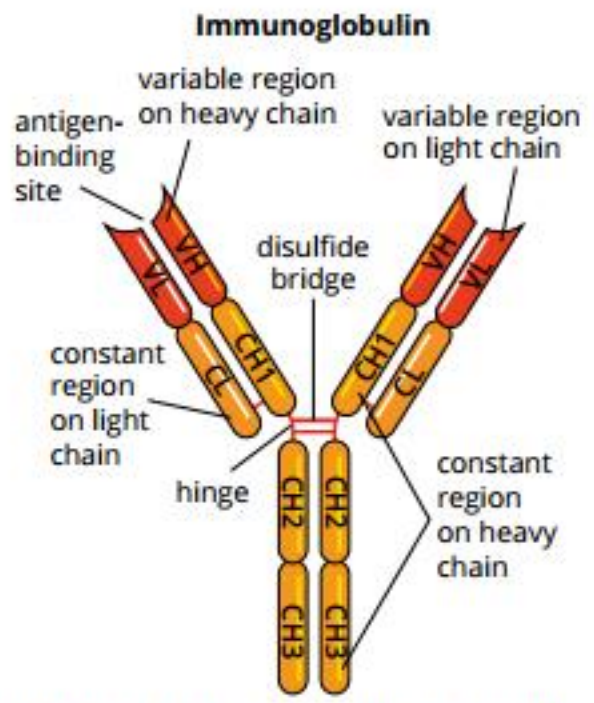


FIGURE 8.2.2 The structure of a basic unit of an antibody. Antibodies have two long heavy (H) chains and two short light (L) chains. Both heavy and light chains have variable (V) and constant (C) regions. Naturally produced antibodies consist of two identical variable regions that are specific for a particular antigen. The constant region can bind to and initiate other immune components, such as the complement proteins.

You will recall from Module 8.1 that that if a vertebrate's first line of defence is breached by a pathogen, that pathogen is met with a non-specific innate immune response. If this innate immune response is not successful in eliminating the invader, vertebrates also have an additional adaptive immune response to pathogens.

There are two distinguishing features of adaptive immunity. The adaptive immune response is specific. On recognising a specific foreign antigen, cells of the adaptive immune system implement an array of defensive mechanisms. The adaptive immune response involves immunological memory. This means that after the first encounter with an antigen, subsequent encounters result in faster and stronger responses.

Adaptive immunity involves antibodies, **B lymphocytes**, and a number of different types of **T lymphocytes**. Figure 8.2.1 shows a B lymphocyte. B lymphocytes and T lymphocytes are types of white blood cells that are important in the adaptive immune response. B lymphocytes produce antibodies, which are proteins that bind to specific antigen molecules.

ANTIBODIES

Antibodies, also known as **immunoglobulins (Ig)**, are produced by B lymphocytes and released into the blood and lymph fluid. When an organism is first exposed to a new foreign antigen, it reverse engineers the protein structure of the antigen. It then changes one or some of many immunoglobulin genes in the B lymphocytes, so it is then able to produce a protein with complementary binding sites (an antibody) to the foreign antigen. Each antibody is specific to only one type of antigen. Pathogens can have many different antigens so each pathogen can stimulate the production of many different types of antibodies.

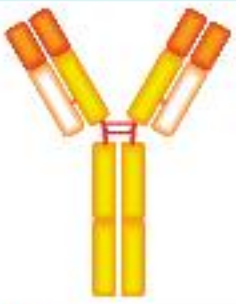
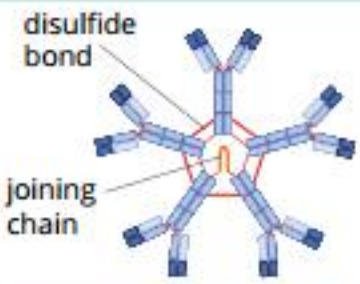
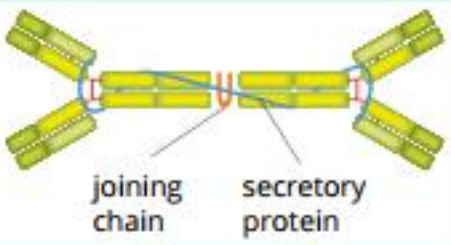
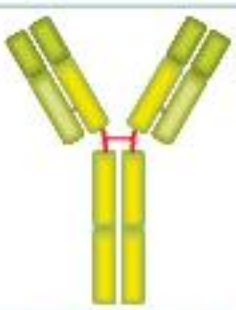
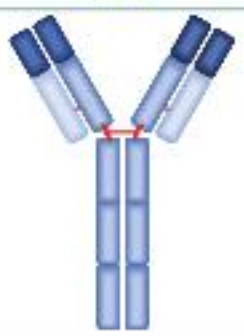
Antibody structure

The basic unit of an antibody molecule is a Y-shaped protein, formed by four polypeptide chains: two long **heavy chains** and two short **light chains**, as shown in Figure 8.2.2. The amino acid sequences that form the top of the 'arms' of the Y-shaped antibody are known as the **variable regions**. The variation of these variable regions allows antibodies to bind to different antigens. The two variable regions are identical antigen-binding sites and attach to identical antigens. The single 'stem' of the Y-shaped antibody is a conserved sequence in all antibodies and is called the **constant region**. The constant region recruits other components of the immune system.

Mammals have five main classes of antibody molecules with different structures and functions. Antibodies may act singly (monomers), in pairs (dimers) or in groups of five (pentamers). Antibodies IgG, IgE and IgD are all monomers but differ in the structure of their constant regions. IgA is a dimer and IgM is a pentamer.

Different classes of antibodies are also found in different body fluids and tissues. The different classes of antibody also play different but overlapping roles in immunity. For example, both IgG and IgA have a role in protecting newborn babies. IgG can cross the placenta and IgA is found in breast milk so that as levels of IgG from the mother decline, the newborn still has some protection. Table 8.2.1 summarises the roles and structure of the five classes of antibody and Figure 8.2.3 illustrates a three-dimensional visualisation of the structure of antibodies.

TABLE 8.2.1 Structure and function of mammalian immunoglobulins

Class	Half-life in serum (days)	Presence	Functions	Structure
IgG	21	Blood, lymph and extracellular fluid; most circulating antibodies (>80%); crosses placenta	agglutination, complement activation	
IgM	10	Blood and lymph; produced early in infection response	agglutination, complement activation	
IgA	6	Secretions such as tears, saliva and milk	mucosal immunity	
IgD	3	Blood and lymph; mostly present on B lymphocyte surfaces; small amount in circulation; binds to basophils and mast cells	functions not well understood; possible role in regulating innate immune responses	
IgE	2	Blood and lymph; attaches to mast cells	involved in allergic reactions	

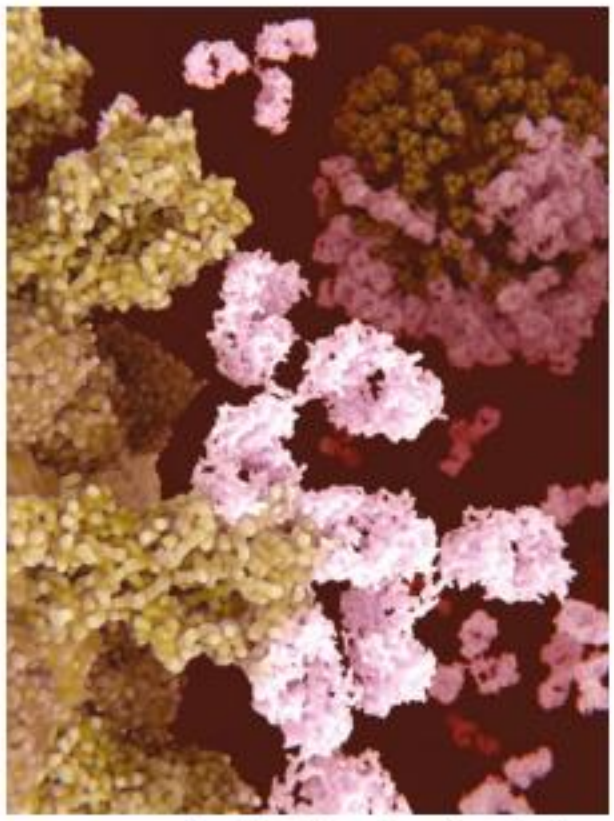


FIGURE 8.2.3 A digital illustration of antibodies (pink) binding to specific antigens on the surface of influenza virions.

Antibody functions

Antibodies do not directly destroy pathogens but carry out several important mechanisms to interfere with the function of the pathogen. The major functions of antibodies are:

- neutralisation of toxins—antibodies bind to toxins produced by bacteria or other organisms, blocking the action of the toxin
- neutralisation of pathogens—antibodies bind to antigens on the surface of the pathogen that are required for entry into host cells, thus preventing pathogen invasion of host cells
- activation of the complement cascade
- **agglutination**—antibodies bind to antigens on the surface of cells and form antigen–antibody complexes, which activate phagocytes and the complement cascade, leading to antigen/cell destruction. Agglutination is an advantage because it immobilises that pathogen and facilitates the phagocytosis of large numbers of pathogen particles at the same time
- **precipitation**—antibodies bind to soluble antigens, causing them to become insoluble and precipitate out of solution.

The major functions of antibodies are summarised in Figure 8.2.4.

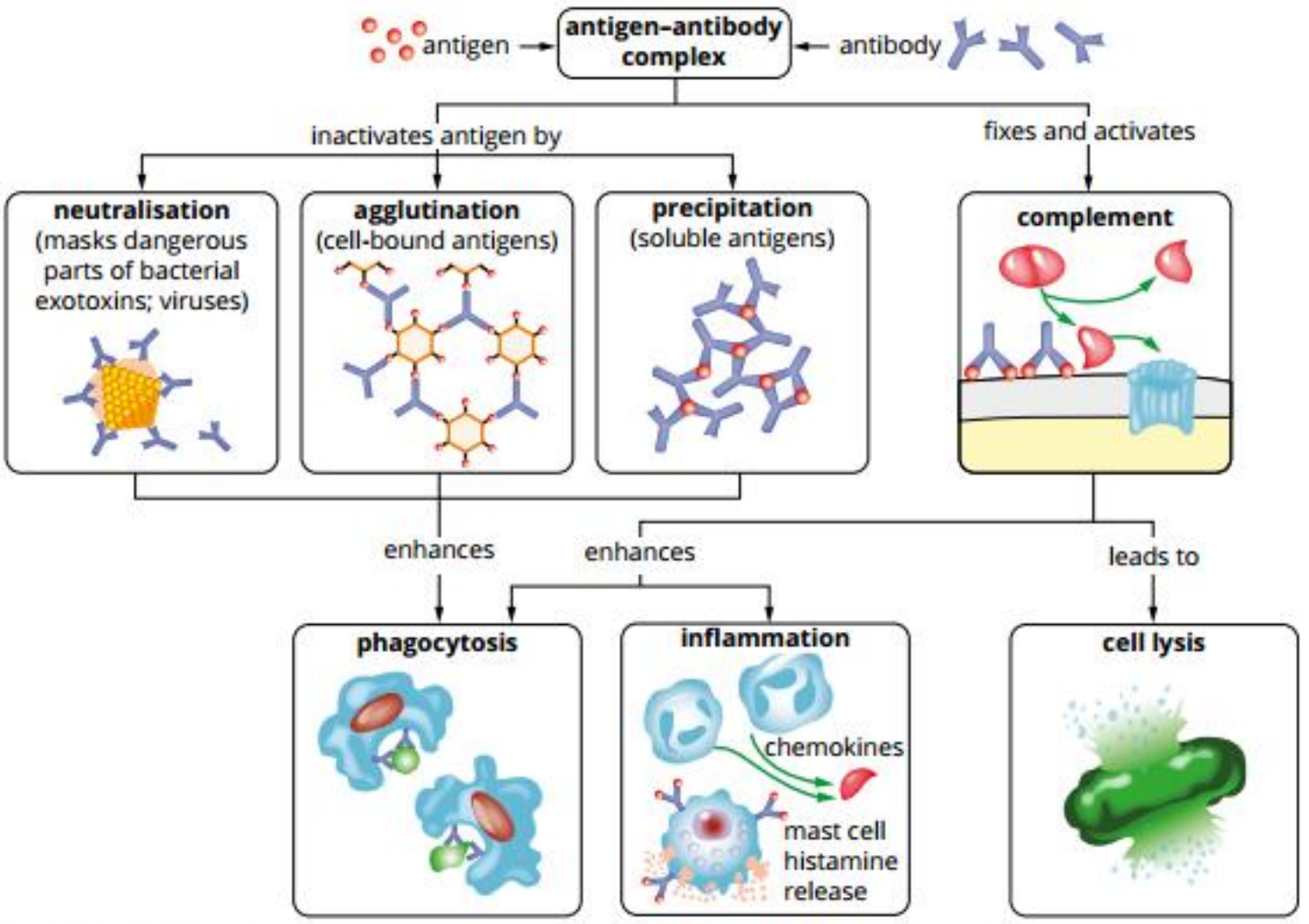


FIGURE 8.2.4 Antibodies function in a number of different ways to help eliminate pathogens.

MECHANISMS OF ADAPTIVE IMMUNE RESPONSES

A large part of the specificity of the immune response is achieved by having cells with specific receptors. Each lymphocyte has a different receptor. Receptor production is controlled by the MHC genes. Because these genes are constantly rearranged, there are many different receptors, each on a different lymphocyte, and each one is specific to a particular antigen. The shape of a receptor matches a molecule found on the specific pathogen.

The cells of the adaptive immune response are lymphocytes. Lymphocytes are white blood cells, which are formed in the bone marrow of long bones. These lymphocytes include B lymphocytes and a variety of T lymphocytes.

There are two mechanisms of adaptive immunity:

- **cell-mediated immunity**, which involves the action of T lymphocytes and phagocytes
- **humoral immunity**, in which macromolecules, such as complement proteins, and antibodies produced by B lymphocytes, are secreted into the extracellular fluid.

The processes of the adaptive response are mediated by cytokines, the signalling molecules of the immune system. One important group of cytokines involved is the **interleukins**.

Cells of the adaptive immune response

B lymphocytes originate and begin differentiating in the bone marrow and complete their maturation in the peripheral lymphoid organs and tissues, especially lymph nodes. B lymphocytes which would react to self-antigens are either suppressed or destroyed early in the development process in the marrow. During the maturation process, most B lymphocytes become antibody producing **plasma cells**, but some become **memory B lymphocytes**. Figure 8.2.5 illustrates the structures of the lymphatic system. At any one time, there are billions of B lymphocytes circulating in the blood.

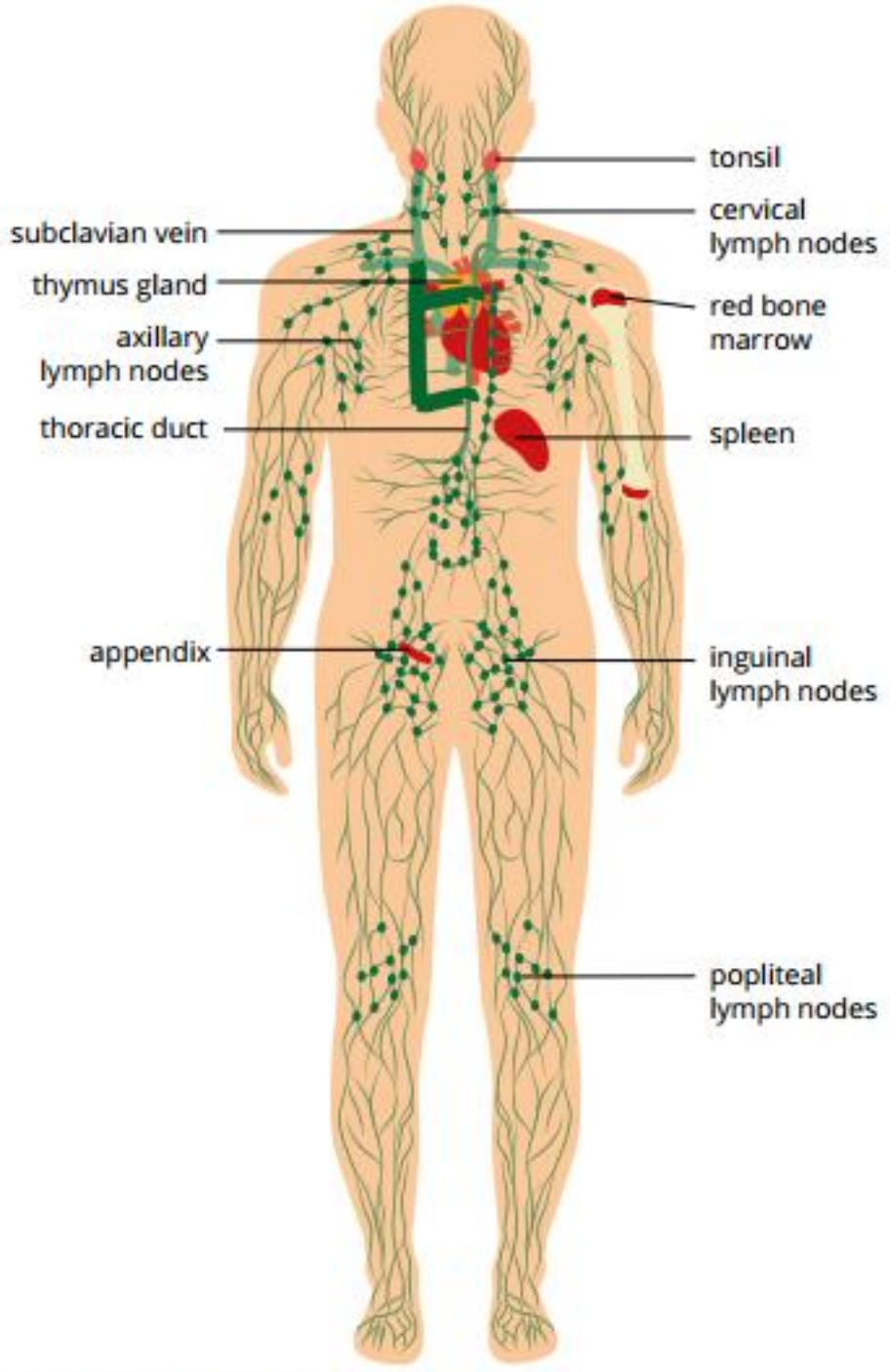


FIGURE 8.2.5 The main structures of the lymphatic system

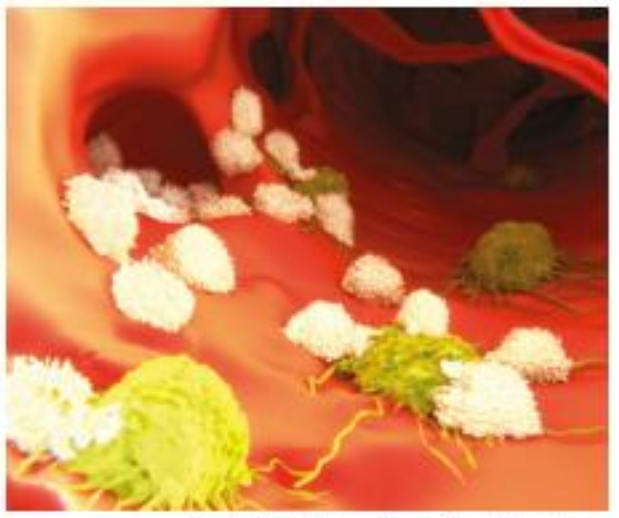


FIGURE 8.2.6 A digital illustration of cytotoxic (white) T lymphocytes attacking migrating cancer cells (yellow)

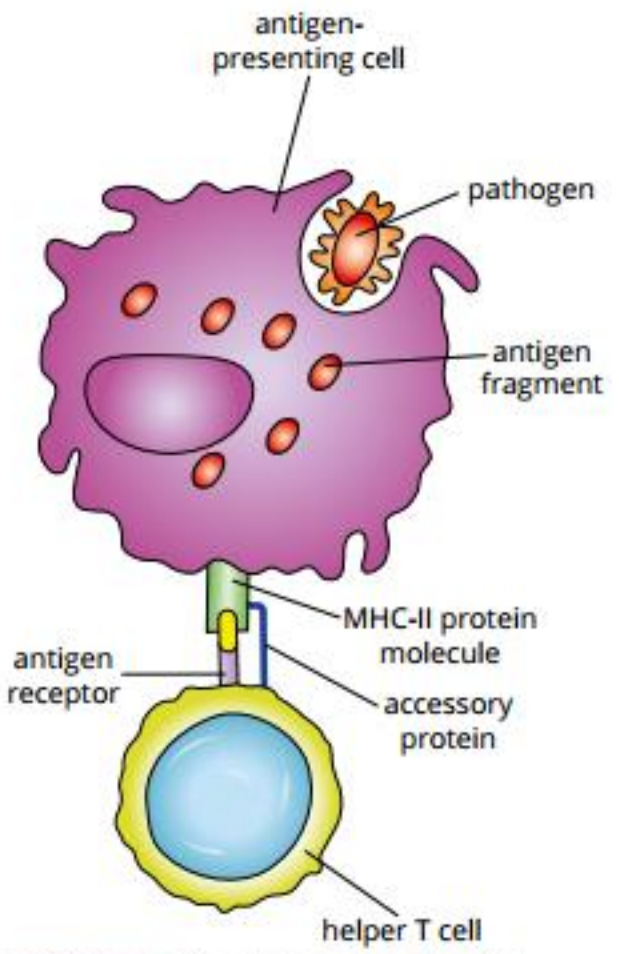


FIGURE 8.2.7 An antigen-presenting cell presenting an antigen to a T_H cell attached to its MHC-II marker. The T_H cell has a receptor with a shape complementary to the antigen and it becomes activated.

T lymphocytes are made in the bone marrow and then move to the thymus gland where they mature. Any T lymphocyte which has a receptor which would bind to a self-cell is destroyed in the thymus gland, thereby maintaining self-tolerance.

There are a variety of T lymphocytes, and each has a specific role to play in the immune response.

- Helper T lymphocytes (T_H) do not directly kill pathogens, but they assist the immune response by secreting cytokines that promote inflammation and activate macrophages and B lymphocytes.
- Cytotoxic T lymphocytes (T_C) recognise and kill foreign, infected or abnormal host cells by releasing toxic compounds (Figure 8.2.6).
- **Memory T lymphocytes** differentiate into memory T lymphocytes that are antigen-specific. The memory T lymphocytes persist after the infection is resolved, to ensure a prompt response should the same pathogen re-infect the organism.
- **Regulatory T lymphocytes** (also known as suppressor T cells or T_{REGS}) suppress the immune response after the infection has been defeated.

Initial adaptive immune response

Adaptive immunity relies on antigen recognition. APCs engulf, process and display antigens to B and T lymphocytes, which are part of the adaptive response. The APCs display the antigens attached to MHC-I proteins to cytotoxic T lymphocytes or the antigens attached to MHC-II proteins to T helper lymphocytes.

When a T_H lymphocyte meets an antigen-presenting cell with an antigen that matches its receptor, that T_H cell becomes activated. This is shown in Figure 8.2.7.

Once the T_H cell is activated, it either activates a B lymphocyte, starting the humoral pathway, or activates a T_C lymphocyte and the cell-mediated pathway.

Humoral immunity targets antigens in the tissue fluids or attached to cells, and involves B plasma cells, whereas cell-mediated immunity focuses on infected or defective cells and involves T_C cells. Most lymphocytes are similar in appearance, but have different functions, as shown Figure 8.2.8.

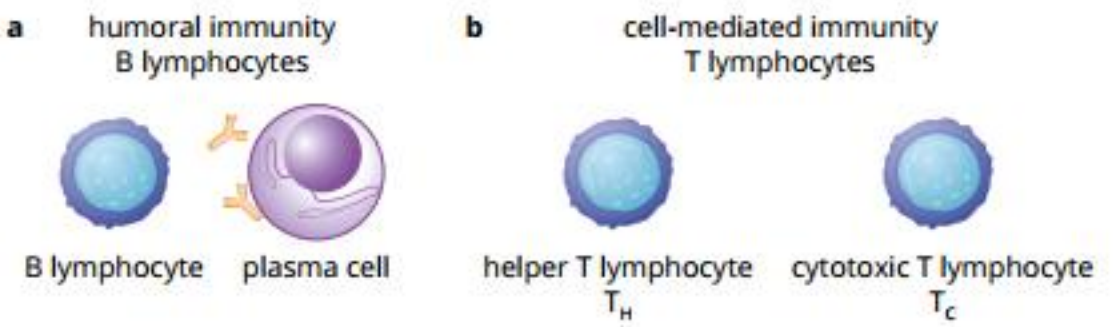


FIGURE 8.2.8 (a) B lymphocytes are involved in humoral or antibody-mediated immunity. They originate in bone marrow and mature in the peripheral lymphoid organs and tissues. (b) T lymphocytes are involved in cell-mediated immunity. They originate in bone marrow and mature in the thymus. Except for plasma cells, the different types of lymphocytes look very similar under a microscope. The only way to identify them is by their different surface proteins.

Humoral immunity

Humoral immunity involves proteins present in body fluids such as blood, tissue fluid and lymph. It is mediated by B lymphocytes.

Both T lymphocytes and the antibodies produced by B lymphocytes can recognise the presence of non-self antigens. The receptors on B lymphocytes are membrane-bound antibodies that recognise free antigens or antigens that are on the surface of a pathogen. Antibodies can also be secreted by the B lymphocytes. Figure 8.2.9 illustrates the different types of antigen receptors.

i The term 'humoral' derives from medieval times, when body fluids were called humours.

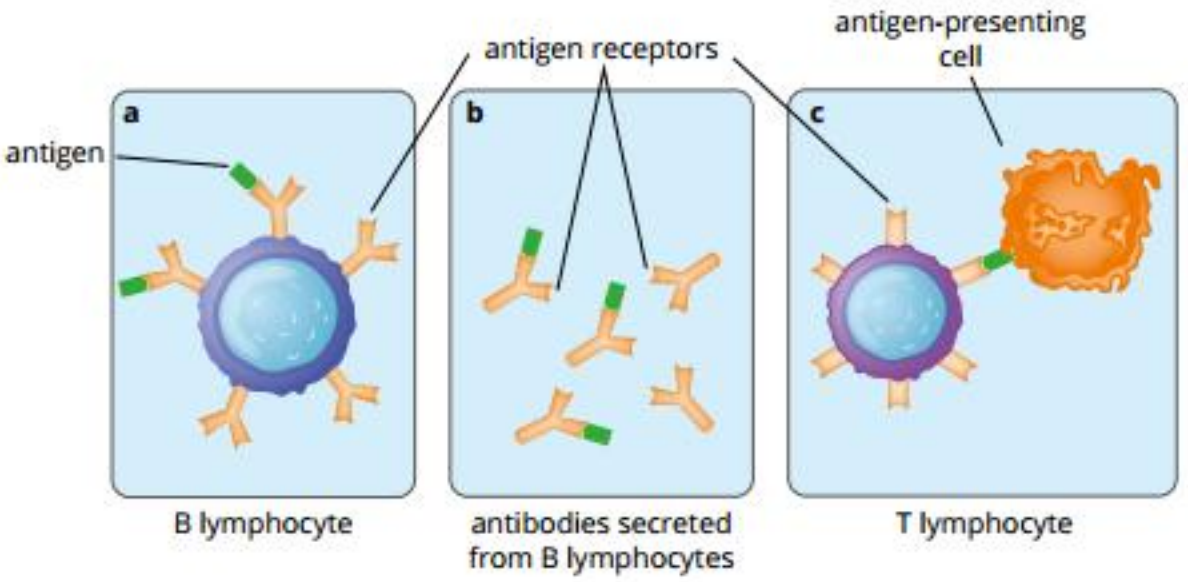


FIGURE 8.2.9 Antigen receptors on immune cells. Antibodies are antigen receptors that may be (a) bound to the cell membrane of B lymphocytes or (b) secreted from B lymphocytes. (c) T lymphocytes have their own type of receptors that recognise antigens presented by specialised antigen-presenting cells.

When a **B lymphocyte** meets and binds to a specific antigen, it becomes 'primed'. Then when the B lymphocyte meets a T_H cell, which has the appropriate receptor, and has been activated by being presented with the same antigen by an APC, the B lymphocyte starts to proliferate. Cytokines released by the T_H lymphocytes regulate this process.

The identification and activation of particular lymphocyte with the specific receptor for a particular antigen is called **clonal selection**. Once a lymphocyte has been selected, it proliferates, creating clones of the initial lymphocyte with the specific receptor for the antigen. This process is called **clonal expansion**.

Following activation, B lymphocytes proliferate rapidly to form many new clones. These clones differentiate to form either plasma cells or memory B lymphocytes. Plasma cells are essentially 'factories' specialising in antibody production, as shown in Figure 8.2.10. The antibodies produced are specific to the antigen that activated the B lymphocyte. A plasma cell can produce 2000–10 000 antibody molecules per second.

A lymphocyte that is mature but has not come in contact with the antigen specific to its receptor is described as 'naïve'.

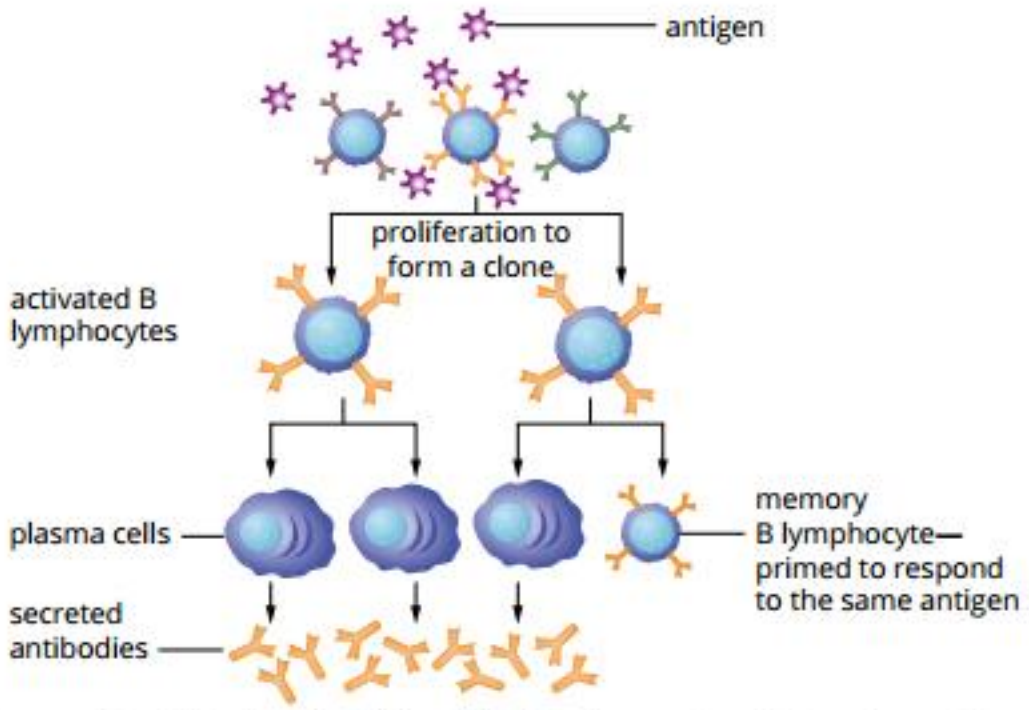


FIGURE 8.2.10 Many B lymphocytes differentiate into plasma cells, which produce and secrete antibodies for immune protection. Others become memory B lymphocytes and are retained in lymph nodes. Helper T lymphocytes are often involved in activating B lymphocytes to produce antibodies.

Memory B lymphocytes are one source of immunological memory. They remain in lymphoid tissues for long periods (even for the lifetime of the animal) and are responsible for the immunity that often follows infection or vaccination. These cells can divide rapidly and give rise to new plasma cells if secondary exposure to the antigen occurs.

The millions of antibodies produced by plasma cells bind to the antigens attached to the pathogen or molecules released by the pathogen and thereby inactivate them (Figure 8.2.11).

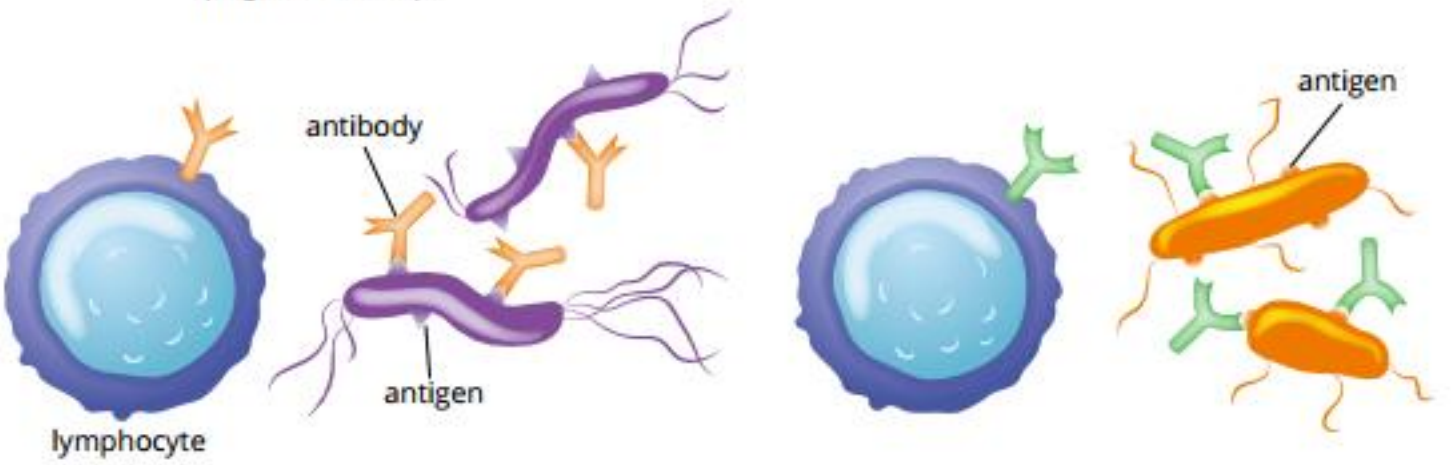


FIGURE 8.2.11 Antibodies specific to a foreign antigen bind to it, helping to eliminate the invading pathogen.

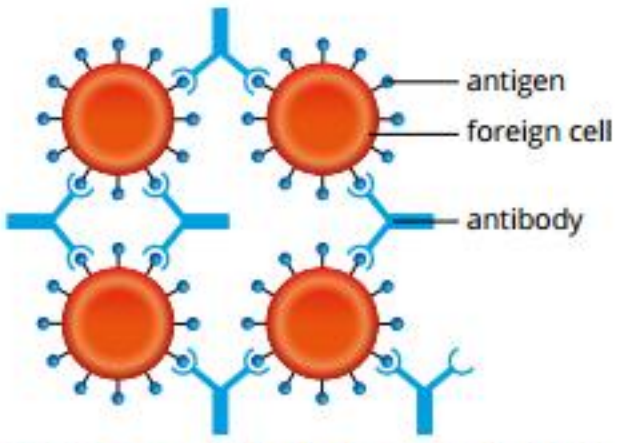


FIGURE 8.2.12 Antibodies have two attachment sites that can attach to two different pathogens, using the same antigen, and link them together (agglutinate), immobilising them and making it easier for phagocytes to engulf the pathogens.

Antibodies can also form cross-links between pathogens, such as bacteria, clumping them together and making it easier for phagocytes to engulf them. This is shown in Figure 8.2.12.

Clonal selection and expansion theory

An almost infinite number of different antigens exist, and the immune system can produce lymphocytes specific to each antigen upon exposure. The clonal selection theory explains how lymphocytes are able to produce a large number of antibodies specific to an antigen.

When B and T lymphocytes form, each has a receptor that reacts to a single antigen. The clonal selection theory states that a specific antigen only activates a lymphocyte with a receptor that specifically recognises that antigen. Recognition occurs when a part of the antigen, called the **epitope**, attaches to the receptor. This attachment occurs because the receptor and epitope are complementary in shape. Once activated, the B lymphocyte interacts with a T_H lymphocyte, which releases cytokines. This triggers the proliferation and activation of the B lymphocyte into a clone of millions of effector cells dedicated to eliminating the specific antigen that stimulated the immune response. These effector cells are called plasma cells. Figure 8.2.13 illustrates this process.

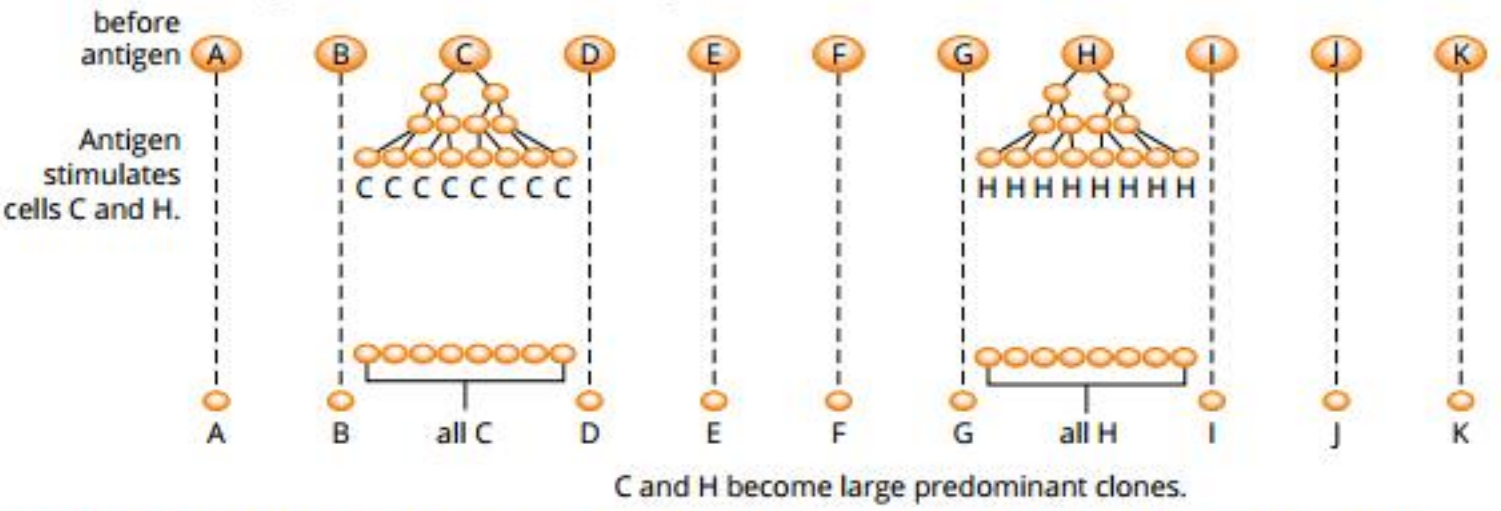


FIGURE 8.2.13 The clonal selection theory and expansion. Lymphocytes that encounter or interact with an antigen, such as lymphocytes C and H, begin to proliferate. This increases the number of lymphocytes with identical receptors, or clones, for the specific antigen that was first encountered. Plasma cells produce antibodies that have antigen binding sites the same shape as the receptor that first recognised the antigen. Each plasma cell can produce only one type of antibody.

Cell-mediated immunity

Unlike humoral immunity, which involves B lymphocytes, cell-mediated immunity is regulated by T lymphocytes. Depending on their function, T lymphocytes are classified as helper, cytotoxic, memory and regulatory T lymphocytes.

The response is mediated by the **T cell receptors** (TCRs). TCRs are central to the function of T lymphocytes in the adaptive immune response (remember that T lymphocytes are also known as T cells). TCRs are made up of two polypeptide chains. Like antibodies, TCRs have a variable and constant region, as shown in Figure 8.2.14. Unlike antibodies, which have two antigen-binding sites, TCRs have only one antigen-binding site.

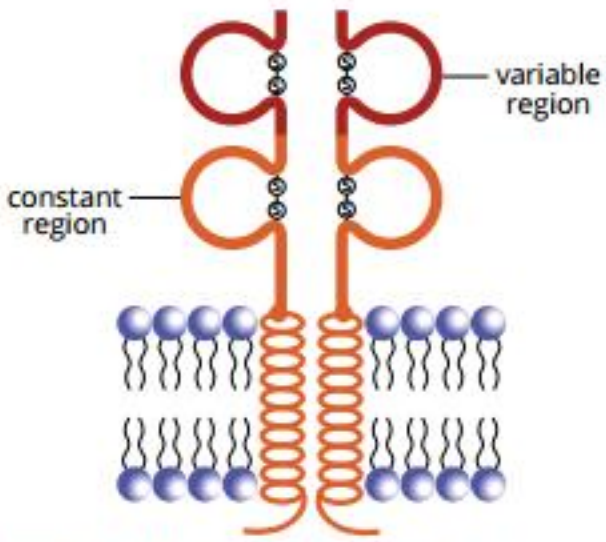


FIGURE 8.2.14 The structure of the T cell receptor (TCR), which is found on helper T and cytotoxic T lymphocytes and binds to fragments of antigen

TCRs do not bind to antigens on pathogens as B lymphocyte receptors do; instead, they bind to fragments of antigens that are displayed or presented on the surface of APCs such as dendritic cells and macrophages. Receptor binding triggers signal transduction in the T lymphocyte, resulting in proliferation, cytokine release and activation of cytotoxic function.

T lymphocytes check the antigens of cells they come into contact within the body, differentiating between cells that belong to the organism (self) and cells that are foreign (non-self). Remember that during their development, lymphocytes that react to self-antigens are normally destroyed. This inability of lymphocytes to respond to self-antigens is known as self-tolerance.

All nucleated cells have surface proteins that present peptide antigens of the proteins being synthesised in that cell. These antigens are presented to cytotoxic T lymphocytes by MHC-I molecules. APCs are specialised for presenting antigens. When an APC engulfs a pathogen, the antigens of the pathogen are broken into small peptides in the cell. These antigen fragments bind to MHC-II molecules inside the cell. The antigen–MHC-II complexes then move to the cell surface to present the antigens to helper T lymphocytes. The TCRs on the helper T lymphocytes recognise the antigen–MHC-II complex. The bone marrow produces millions of different immature T lymphocytes and each has a TCR that is a unique shape that is complementary to only one antigen. Joining the TCR with its unique antigen attached to the MHC marker on the APC initiates signal transduction within the immature T lymphocyte, which then proliferates. The newly formed T lymphocytes differentiate to become helper T, cytotoxic T, memory T or regulatory T lymphocytes, all of which possess the unique receptor that is complementary to the antigen presented by the APC.

The newly formed helper T lymphocytes then activate the cytotoxic T lymphocytes. The whole process is regulated by cytokines. This process is shown in Figure 8.2.15.

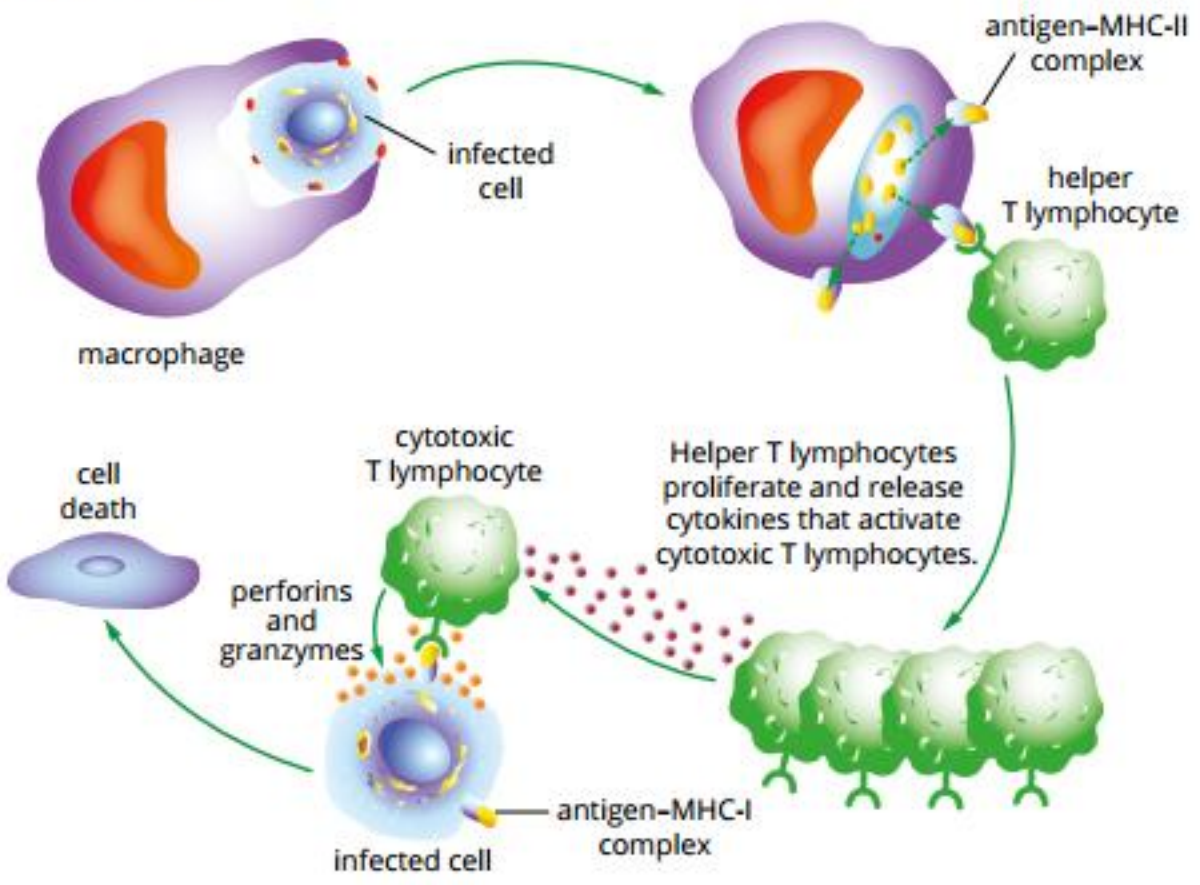


FIGURE 8.2.15 T lymphocytes are activated in cell-mediated immunity. Infected cells are detected and destroyed by cytotoxic T lymphocytes.

The processes of the adaptive immune response are illustrated together in Figure 8.2.16.

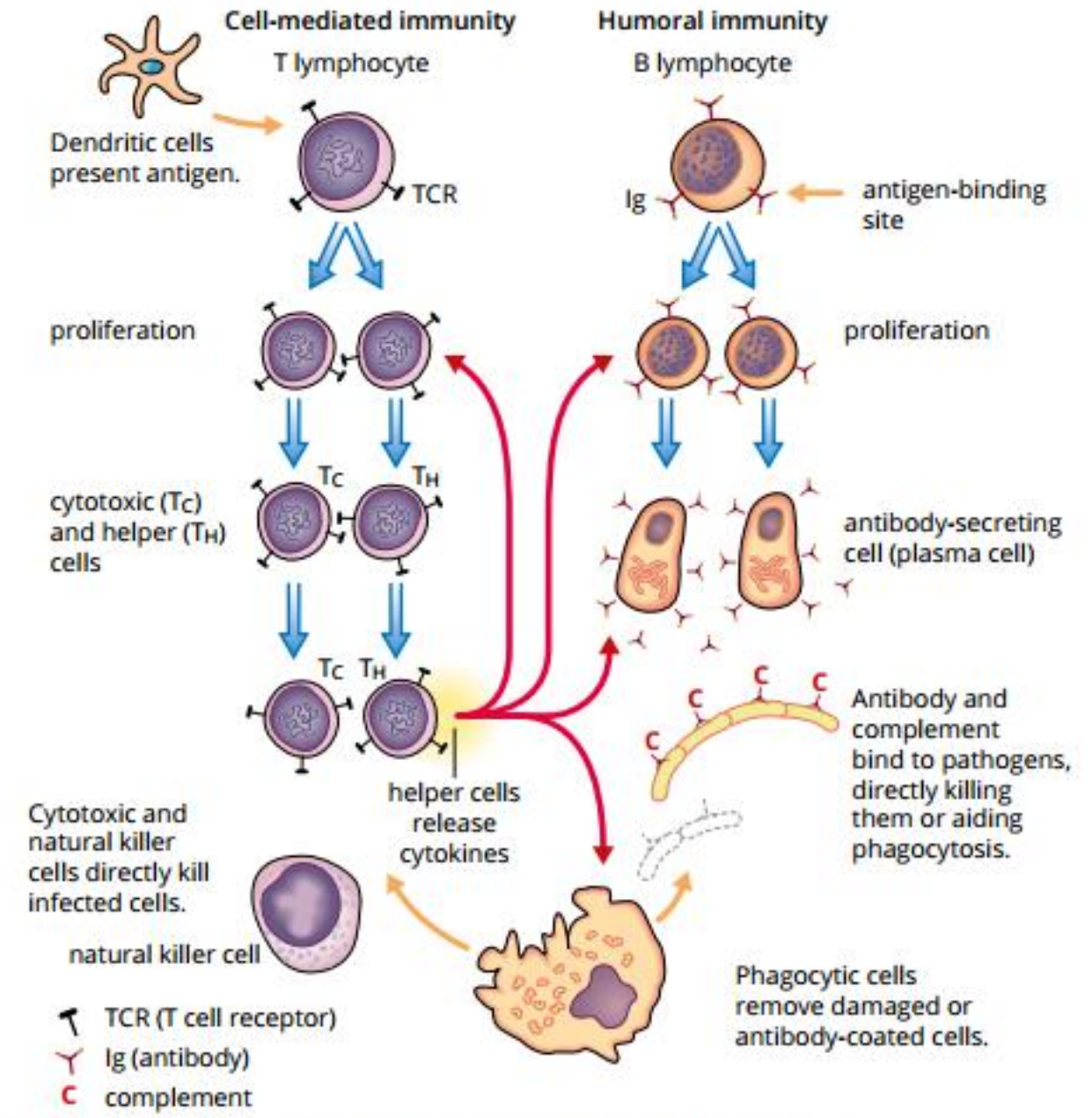


FIGURE 8.2.16 A summary of cell-mediated immunity and humoral responses

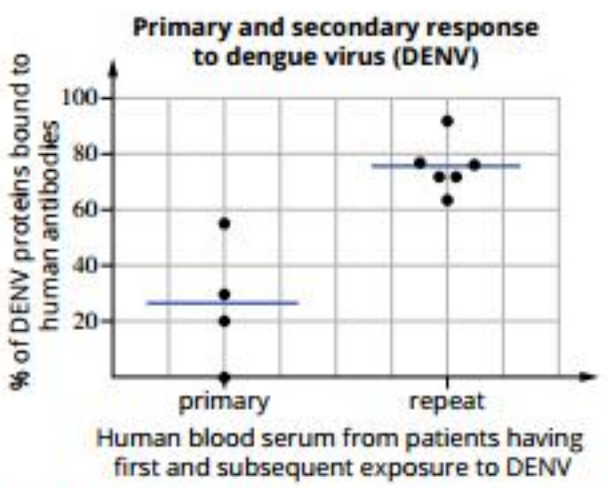


FIGURE 8.2.17 Antibody levels after infection by dengue fever virus (DENV) on initial exposure and subsequent exposure. The secondary response to dengue virus antigens is stronger than the primary response, as shown by the greater binding between human antibodies and the dengue virus antigens.

Immunological memory

The response arising from the first encounter of a T or B lymphocyte with a specific antigen is known as the **primary immune response** (Figure 8.2.17). After the initial exposure, some B and T lymphocytes form B and T memory lymphocytes. IgM antibodies are the predominant antibodies produced in a primary response.

Once formed, lymphocytes constantly circulate between lymphoid tissues, especially lymph nodes, and the bloodstream. Each lymphocyte moves from the lymphoid tissue into the blood and back again about 50 times per day. This maximises the chance of a particular lymphocyte meeting and recognising its complementary antigen if that antigen enters the body.

The response arising from subsequent encounters with the same antigen is known as the **secondary immune response**. Lymphocyte proliferation and production of antibodies occurs much more quickly during the secondary immune response because the existing memory lymphocytes, which were produced during the first encounter and remain for months or years, allow faster proliferation of the required lymphocytes (those with the receptor specific to the antigen). At each subsequent infection following the first, more memory cells are rapidly produced. IgG antibodies are the predominant antibodies produced in the secondary response.

A secondary antibody response is stronger than the primary response. This has been shown by many experiments. Figure 8.2.17 shows the results of patients' antibody levels after infection by dengue virus. Some patients were experiencing the virus for the first time while others were having secondary exposure. Blood samples were taken from the patients and their blood serum was incubated with dengue virus particles and the percentage of cross-linking between the antibodies and the virus particles was determined. Figure 8.2.18 summarises the primary and secondary immune responses. You will learn more about the immune response in Module 8.3.

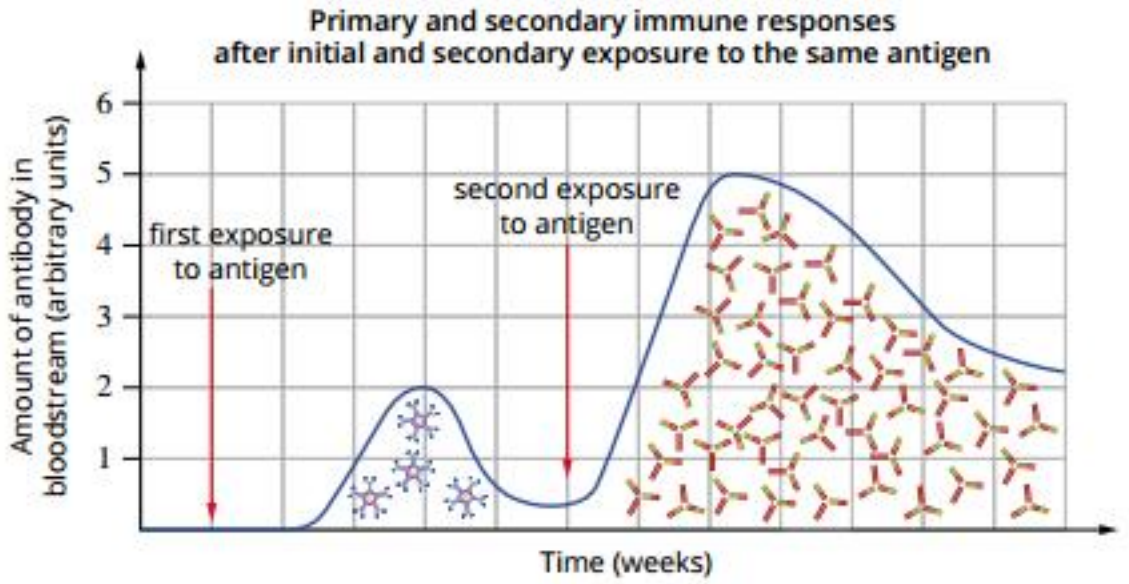


FIGURE 8.2.18 Primary and secondary immune responses after initial and secondary exposure to the same antigen

Memory lymphocytes

Memory lymphocytes are made during any stimulation of the immune system, but the number of memory cells formed can vary in response to different antigens. Memory cells of both kinds usually have long lives, with some memory cells remaining throughout a person's life. There is some variation in this and for some diseases immunological memory becomes depleted over time.

The antigen binding sites of memory lymphocytes have a higher affinity for their corresponding antigen and thus react faster than naïve B and T lymphocytes. Naïve B or T lymphocytes with a suitable antigen receptor may be very limited in number. Activation is a multi-step process and may take some time. The higher the number of memory cells and greater their affinity for their corresponding antigen means that the secondary and subsequent responses are faster and produce a greater number of antibodies.

Immunological memory is very effective against some pathogens because a strong immune response is mounted, and many memory cells are made but is less effective against others due to a weaker response occurring. The reasons for this variation in response are not well understood. Moreover, immunological memory becomes less effective if the pathogen mutates rapidly so its antigens become very different and thus a new primary response is required.

8.2 Review

SUMMARY

- Vertebrate immune responses are specific and involve memory cells.
- Specific defence is provided by an adaptive immune system.
- Specific immune responses involve lymphocytes, called B and T lymphocytes, which are made in the bone marrow.
- Adaptive immunity is divided into the humoral and cell-mediated responses.
- The adaptive response is initiated when phagocytes present antigens to helper T cells.
- The humoral response is enacted by B lymphocytes, which undergo clonal expansion and produce specific antibodies after being activated by T_H cells.
- Antibodies are proteins that cause neutralisation, agglutination and precipitations of pathogens and their toxins.
- The cell-mediated response is enacted by T lymphocytes against cancer cells, transplanted cells and cells infected by viruses.
- Once activated both B lymphocytes and T lymphocytes produce memory cells during clonal expansion during the initial infection.
- Memory B and T lymphocytes remain in the body for long periods of time and provide a faster, stronger immune response to subsequent infections involving the same antigen/pathogen.
- More memory cells are produced with each subsequent infection.

KEY QUESTIONS

Describe

1 List five functions of antibodies.

2 What are cytokines?

3 How is self-tolerance maintained?

4 Describe the role of memory cells.

Apply

5 Draw an antibody. Label the heavy and light chains, the variable region, the disulfide bridges and the antigen-binding sites.

6 Draw a table of the different types of T lymphocyte and their function.

7 Explain how a naïve B lymphocyte and a B plasma cell differ.

8 Explain why the secondary immune response is faster and greater than the primary immune response.

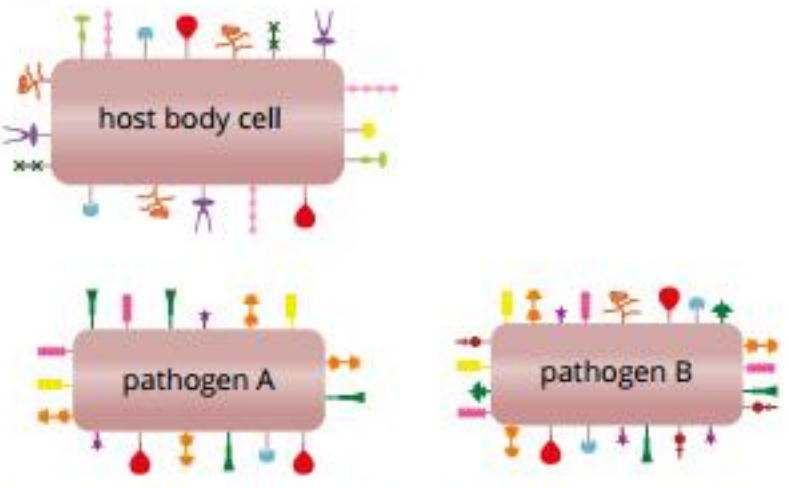
9 Draw a schematic outlining the sequence and mechanisms of:

a humoral immunity

b cell-mediated immunity

Analyse

10 An organism that has cells with self-antigens as shown below is invaded by two different but related pathogens.

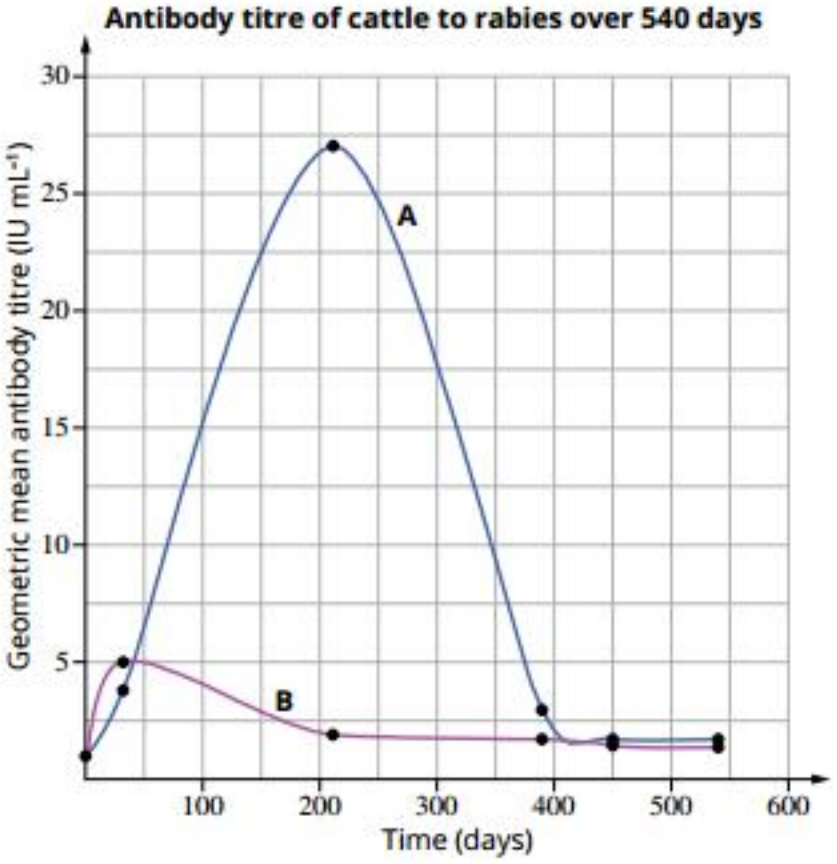


a State the number of different antigens present on the membrane of pathogen A compared to the host cell.

b How many antibodies to pathogen B could the host organism could make?

c Identify which pathogen, A or B, results in the greater variety of antibodies being produced by the host. Justify your answer.

11 An experiment was performed to determine the effectiveness of a vaccine against rabies virus in stimulating the production of antibodies in cattle. Two groups, A and B, of cattle were assessed. Both groups were first exposed to the virus on day 0, then only one of the groups was exposed again on day 120. Antibody titres were measured on days 0, 30, 210, 390, 450 and 540. The results are shown in the following graph.



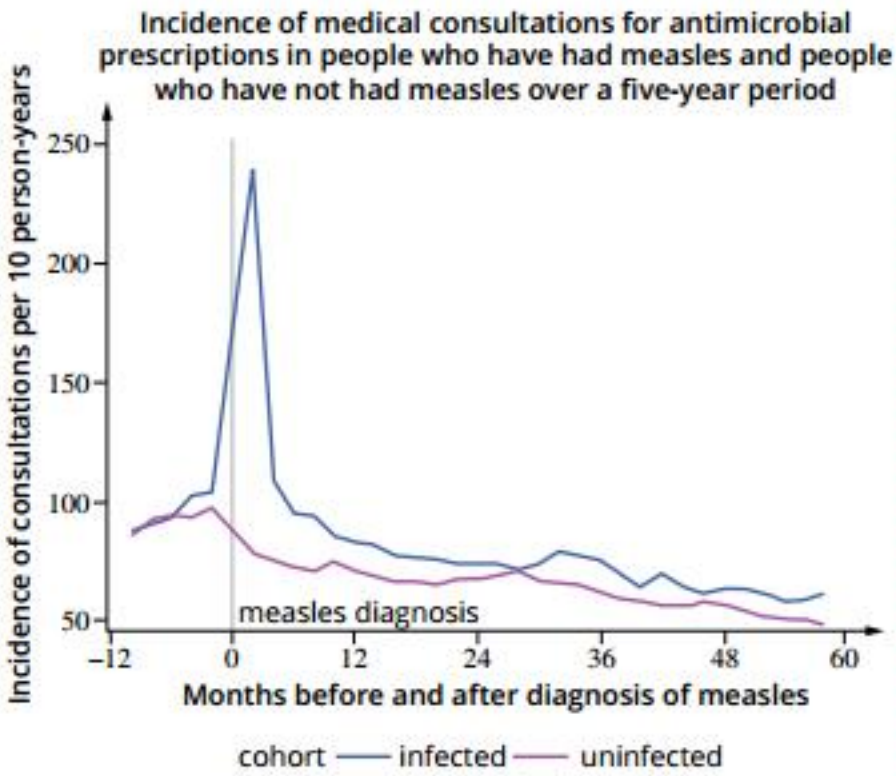
a Deduce which result, A or B, represents the group that was exposed only once. Explain your response.

b **i** Explain why interpolation of the antibody titres for group A between days 30 and 210 would be unreliable.

ii Predict what a scientist who measured the antibody titre of group A would measure if they sampled at day 100.

c Along with increased production of antibodies, T lymphocyte concentration would be expected to rise significantly, especially in the group having two exposures to the pathogen. Explain why.

12 It was suspected that the measles virus damages memory cells thus increasing the chances of contracting other infections in future. During the study two groups were considered. One group was medically diagnosed with measles and the other group never had measles. In order to assess whether long term immunity was compromised, the number of times that each participant in the study attended the doctor and was prescribed an antimicrobial medication such as an antibiotic was tallied. Data was collected for each of the participants over a period of 5 years. The results of the study are shown in the graph below.



a Why would the number of times that a participant was prescribed an antimicrobial medication be a measure of the recipients' immune status?

b Explain whether the data supports the contention that measles damages long-term immunity.

continued over page

8.2 Review *continued*

13 Whooping cough (pertussis) is a life-threatening disease in young children. It is caused by the bacterium, *Bordetella pertussis*. Children in Australia and many parts of the world are vaccinated against it. One study comparing immunity from vaccination versus natural infection was undertaken. Antibody levels and T lymphocyte responses five years post infection or vaccination were determined. All children in the study either had the scheduled three doses of vaccine or caught and recovered from whooping cough during their first 12 months of life. Responses to three different *B. pertussis* antigens (PT, FHA and PRN) were assayed. The results are shown in the table. (n = number of children tested in each group)

***B. pertussis* antigens tested**	**Vaccinated children**		**Naturally infected children**	
	% positive for IgG antibodies (n = 38)	**% with positive lymphocyte (T cell) response (n = 34)**	**% positive for IgG antibodies (n = 21)**	**% with positive lymphocyte (T cell) response (n = 14)**
PT	3	5	5	1
FHA	28	6	15	2
PRN	25	6	5	2

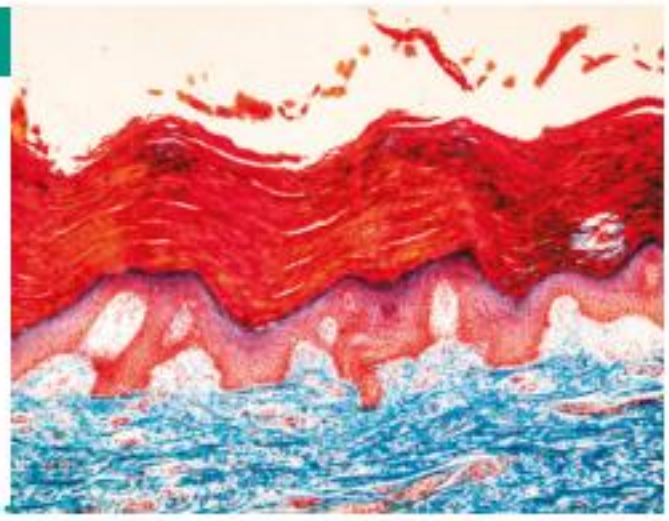
a Compare the responses of the vaccinated with the naturally infected.

b In Australia, children are required to receive a booster which includes pertussis vaccine just before starting school. Using the data, explain why.

8.3 Active and passive immunity

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- understand that passive immunity occurs when individuals acquire antibodies from another organism
- understand that active immunity is obtained when an individual makes memory lymphocytes and their own antibodies
- understand that vaccination has significantly decreased the world burden of disease
- recognise that immunisation rates need to be high in order to provide the community with herd immunity.



In Modules 8.1 and 8.2, you learnt about the physical, chemical and microbiological barriers that help prevent infection, as well as the innate and adaptive immune responses that provide immunity against invading pathogens that bypass these first line defences. In this module, you will learn that immunity can be active or passive, natural or artificial. You will also learn that vaccination (Figure 8.3.1) is an example of artificial active immunity, and about the role of vaccination programs in creating herd immunity and preventing disease.

Immunity is active or passive depending on the origin of the immune response. **Active immunity** results from an organism encountering antigens and then making their own B and T memory lymphocytes. These lymphocytes can last for a long time, even an entire lifetime. **Passive immunity** is generated when antibodies pass from one organism, which made them, to another organism by natural or artificial means.

Active immunity is long term but can take some time to develop. During this time, the pathogen has time to multiply and cause illness. That illness can be so severe that the patient dies before their immune system can respond. However, once the response has occurred and the pathogen defeated, long-term protection is achieved.

Passive immunity is immediate in its action as the antibodies are already formed but no new antibodies will be made so if the initial response is inadequate, illness may follow. Also, passive immunity does not result in the formation of immunological memory, so won't result in protection from future infection.

FIGURE 8.3.1 A baby being vaccinated. The vaccine stimulates an adaptive immune response that protects against infection.

PASSIVE IMMUNITY

Passive immunity is acquired when antibodies made in one organism are transferred to a different individual. Passive immunity generally lasts for less time than active immunity. This is because the transferred antibodies last for a limited time in the organism receiving them. Passive immunity occurs naturally when antibodies pass from mother to fetus through the placenta before birth or through breast milk once the baby is born. **Artificial passive immunity** can be created when antibodies that were made by another person, or an animal such as a horse or rabbit, are injected into an individual.

Natural passive immunity

Natural passive immunity involves the passive transfer of antibodies from mother to fetus, or from mother to breast-feeding baby. These maternal antibodies provide protection to the baby for weeks or months, while the baby's own immune system is developing. The newborn baby relies heavily on these antibodies during the first months of life because its immune system is not fully developed.

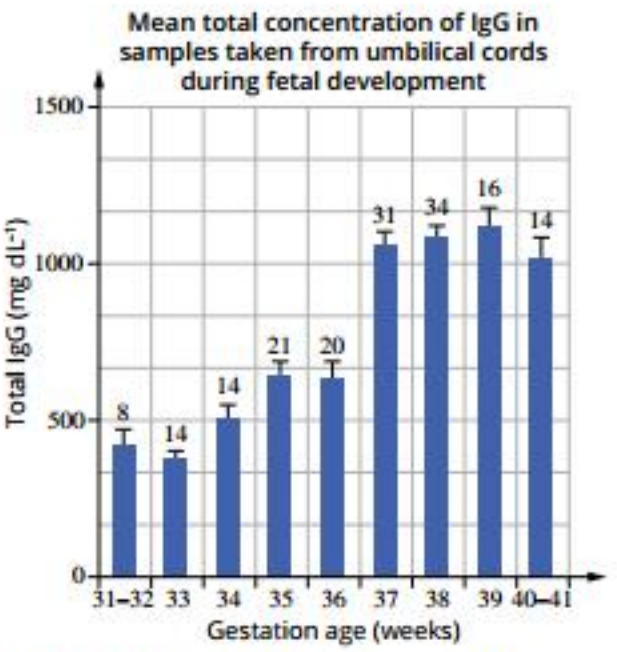


FIGURE 8.3.2 Antibodies are passed from mother to fetus. The numbers above the bars indicate the numbers of samples taken.

FIGURE 8.3.3 Laboratory tests showing an Rh^+ result (top), in which there is D antigen present on red blood cells (third vial from the left), and an Rh^- result (bottom), in which there is a lack of D antigen.

IgG is the only class of antibody that crosses the placenta in significant amounts. The level of IgG crossing is determined by a number of factors, including the IgG concentration in the mother's blood, the gestational age of the fetus, the integrity of the placenta, and the nature of the antigen that caused the generation of the antibody. Figure 8.3.2 shows a typical profile of IgG in a healthy fetus with a healthy mother.

The concentration of placenta-derived maternal IgG antibodies lowers over the first 9 months of the baby's life. However, antibodies continue to be passed to the infant through breast milk. As can be seen in Table 8.3.1, IgG antibody levels are nearly as high in breast milk as in the mother's blood, however, IgA and IgM levels are only about half.

TABLE 8.3.1 Relative antibody levels, determined by enzyme-linked immunosorbent assay (ELISA)

Antibody class	Mean relative maternal concentration in breast milk (±SD)	Mean relative maternal blood serum concentration (±SD)	*P* value
IgG	0.045 ± 0.031	0.052 ± 0.017	0.852
IgA	0.039 ± 0.0075	0.0613 ± 0.0205	0.167
IgM	0.036 ± 0.010	0.052 ± 0.014	0.085

Haemolytic disease of the newborn

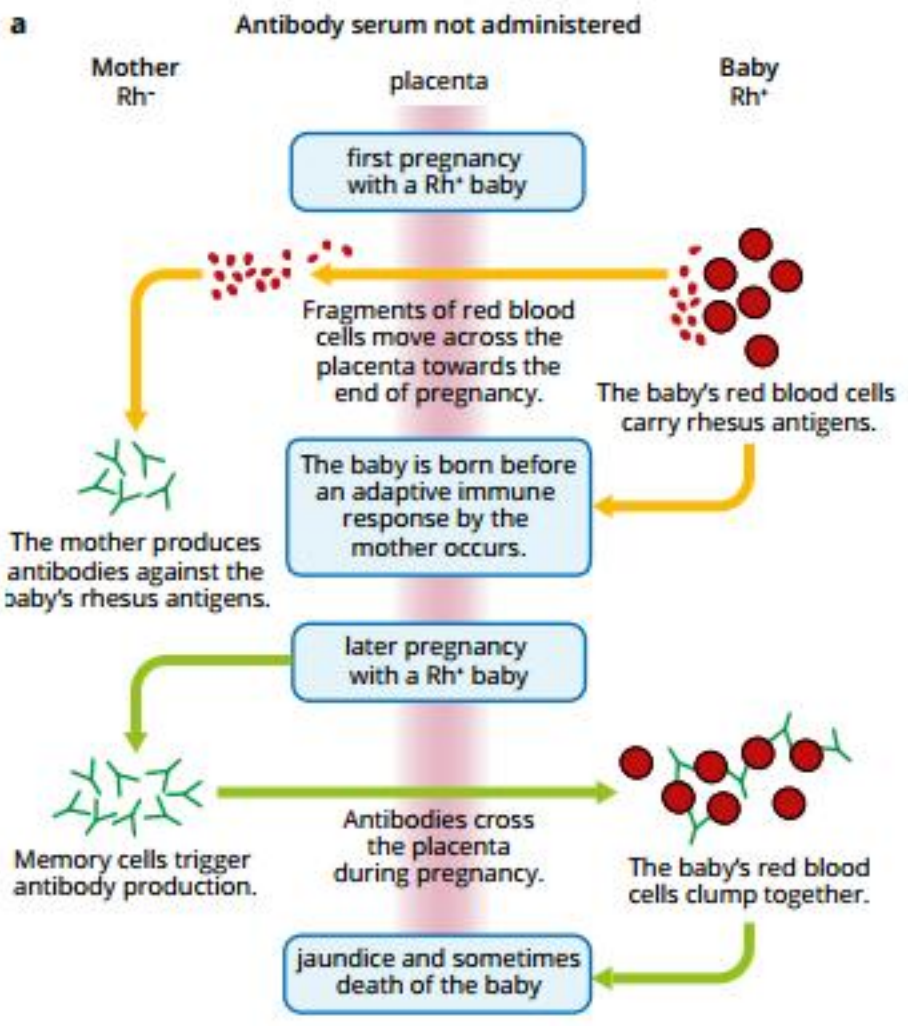
Passive immunity as a result of antibodies crossing the placenta is usually an advantage to the fetus, but sometimes those antibodies can damage the fetus. **Haemolytic disease of the newborn** is caused by maternal antibodies attacking the blood of the fetus.

You will recall from Module 8.1 that blood cells have A and B antigens on their surface, which results in the blood groups A, B, AB and O. More than 100 different antigens have been identified on the surface of blood cells; another one is the Rh, or rhesus, protein, which was first seen in rhesus monkeys. People either have the rhesus antigen (Rh^+) or do not have the protein (Rh^-).

Haemolytic disease of the newborn occurs when a mother's immune system makes antibodies that cross the placenta and attack the red blood cells of her fetus. This occurs when the mother lacks the rhesus antigen (or D antigen) on her red blood cells (Figure 8.3.3). She has rhesus blood type Rh^-. When an Rh^- mother has an Rh^+ baby, the mother is likely to develop an adaptive immune response to the Rh antigen, as fragments of fetal red blood cells cross the placenta during birth (Figure 8.3.4a).

If the same Rh^- woman has another pregnancy with an Rh^+ baby, her memory cells trigger the production of antibodies that cross the placenta and damage the baby's blood cells, causing them to develop haemolytic disease of the newborn (from *haemo*, meaning 'blood', and *lysis*, meaning 'breakdown'). In each subsequent pregnancy with an Rh^+ baby, more memory cells and antibodies are made, and the situation worsens.

To prevent the mother having an adaptive immune response and producing anti-Rh antibodies during her first pregnancy, she is given a dose of anti-Rh antibodies (Figure 8.3.4b). These administered antibodies neutralise any fetal Rh antigens before an adaptive immune response by the mother occurs. The benefits of giving the mother IgG antibodies within 72 hours of the birth have been demonstrated in a number of studies. Table 8.3.2 shows the results of an early study in Canada that proved the benefits of giving IgG antibodies to women who were Rh incompatible with their babies after they gave birth.


a
Antibody serum not administered
Mother
Rh-
placenta
Baby
Rh+
first pregnancy with a Rh+ baby
Fragments of red blood cells move across the placenta towards the end of pregnancy.
The baby's red blood cells carry rhesus antigens.
The baby is born before an adaptive immune response by the mother occurs.
The mother produces antibodies against the baby's rhesus antigens.
later pregnancy with a Rh+ baby
Memory cells trigger antibody production.
Antibodies cross the placenta during pregnancy.
The baby's red blood cells clump together.
jaundice and sometimes death of the baby

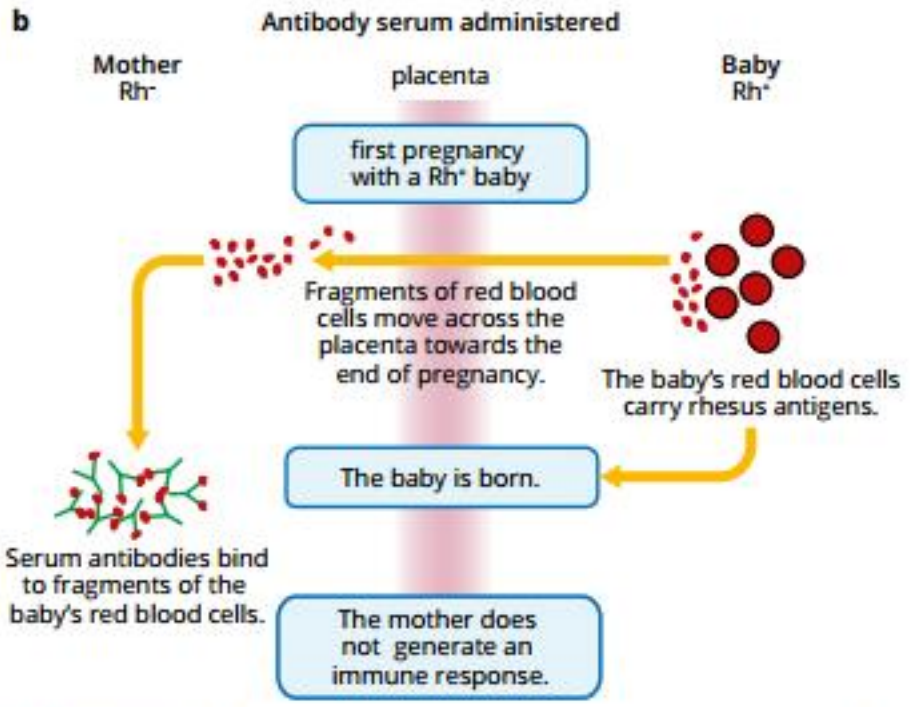

b
Antibody serum administered
Mother
Rh-
placenta
Baby
Rh+
first pregnancy with a Rh+ baby
Fragments of red blood cells move across the placenta towards the end of pregnancy.
The baby's red blood cells carry rhesus antigens.
The baby is born.
Serum antibodies bind to fragments of the baby's red blood cells.
The mother does not generate an immune response.

FIGURE 8.3.4 Haemolytic disease of the newborn can be prevented by artificial passive immunisation. A mother who is Rh^- has a baby who is Rh^+. (a) When red blood cell fragments cross the placenta, the mother has an adaptive immune response that makes memory T and B cells, and antibodies against the Rh antigen (called anti-Rh antibodies). In a later pregnancy with an Rh^+ fetus, memory cells trigger the production of antibodies that can cross the placenta and damage any subsequent Rh^+ fetuses. (b) A dose of anti-Rh antibodies can be administered to the mother to neutralise any fetal Rh antigens before an immune response occurs, protecting the future Rh^+ fetus.

TABLE 8.3.2 Haemolytic disease of the newborn prevention trial (March 1967–January 1968)

	Given Rh IgG after birth	Not given Rh IgG after birth
Rh^- mothers in trial	1216	500
No. mothers with maternally created antibodies 6–9 months after birth	0	36 (7.2%)

Finally, many factors influence whether haemolytic disease of the newborn occurs, including the ABO blood type of the mother and fetus. Some of these factors are not fully understood and are the subject of ongoing research.

Artificial passive immunity

Artificial passive immunity is the process of giving an organism antibodies that have been made by another organism. The process of giving IgG antibodies to mothers of Rh-incompatible babies is one example of artificial passive immunity.

To provide artificial passive immunity, the antibodies are usually given by injection of **antiserum**. Antiserum is serum that contains specific antibodies. **Serum** is the fluid portion of blood that remains after blood cells and material involved in blood clotting have been removed, as illustrated in Figure 8.3.5. Serum contains antibodies, as well as other substances. When the transferred antibodies from the antiserum bind to the antigens on the pathogen or toxin, they form an antigen–antibody complex. This complex inhibits the pathogen or toxin before it does much damage.

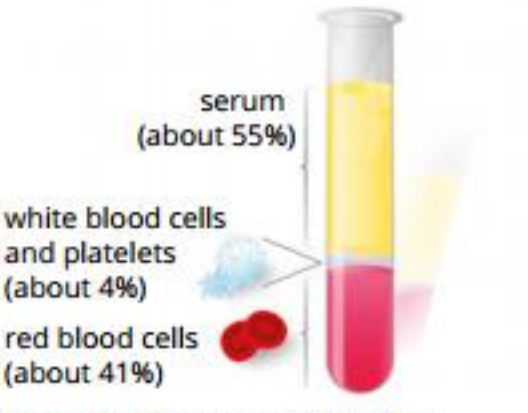


FIGURE 8.3.5 Serum is the fluid portion of blood that remains after blood cells and clotting factors (platelets) have been removed. Antiserum is serum containing specific antibodies injected to treat or protect against disease.

Artificial passive immunisation can be a useful way of treating an infection by a pathogen, or a bite or sting by a venomous animal, when death is likely to occur before the primary immune response has had time to develop. For example, a tetanus antiserum protects against tetanus in at-risk patients, such as those with a deep or dirty puncture wound. The antiserum contains antibodies specific for the toxin, called antitoxins, which bind to the tetanus toxin, inhibiting its function. However, introducing antibodies to contain the threat before the person's adaptive immune response can be mobilised means the protection provided is only temporary, as no immunological memory is formed. In order to achieve long-term immunity, active immunity is required.

ACTIVE IMMUNITY

Active immunity comes from having memory B and T lymphocytes and the ability to make one's own antibodies in response to antigens. Immunological memory is highly specific to the antigens present on a particular pathogen or molecule. Active immunity develops in healthy individuals when foreign antigens are processed by phagocytes and the adaptive response is initiated.

Active immunity can be acquired naturally when someone is infected by a pathogen, becomes ill and then recovers. Active immunity can also be achieved artificially through vaccination. Once active immunity is achieved, it is maintained for a long time, even for the person's lifetime.

Natural active immunity

For most diseases, active immunity is achieved through natural exposure to the disease, illness and recovery, and thus is **natural active immunity**. The large number of pathogen antigens generated during the illness usually results in a strong immune response. For most pathogens, one bout of illness is enough to provide long-term immunity.

Once active immunity is achieved, subsequent exposure to the same antigen results in a secondary response, which is both faster and greater than the initial immune response. Figure 8.3.6 shows the primary and secondary responses to challenge by an antigen. The secondary response is so much larger than the primary response that it needs to be measured on a logarithmic scale (not linear). Also note the time lag before the primary response shows antibody production. This does not occur with the secondary response, which is immediate. The main reason for this difference is the large number of memory cells that were formed during the first exposure to the antigen.

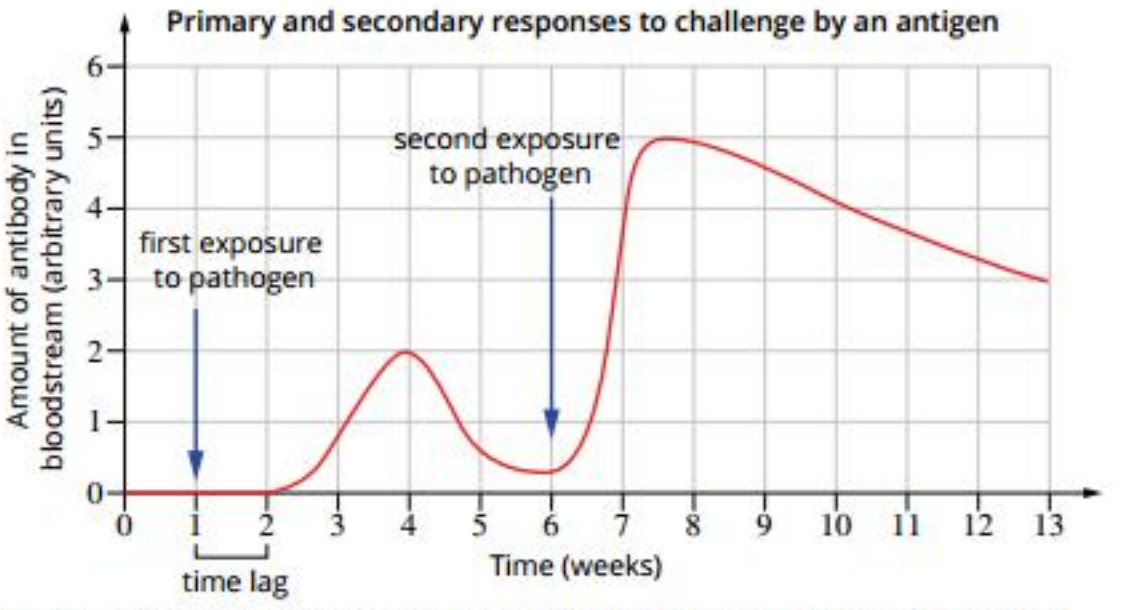


FIGURE 8.3.6 A subsequent infection with the same infectious agent will trigger a secondary immune response. The secondary immune response is faster and stronger than the primary immune response.

Many experiments have demonstrated that antibody levels increase with secondary exposure to pathogens. In one experiment, two groups of newborn mice were used to investigate the development of antibodies to *Leishmania* parasites.

Leishmania parasites are the cause of a serious disease called leishmaniasis. The parasite is spread by an insect vector. As a part of the investigation, mice in the experimental group were exposed to the infected vectors once a week for 5 weeks. Mice in the control group were not exposed to the pathogen. Throughout the experiment, blood samples were taken and tested for IgG antibodies to *Leishmania*. Both groups were exposed to the pathogen at week 27 and their antibody titres were determined one week later. It can be seen in Figure 8.3.7 that the mice that had been previously exposed showed a gradual increase in IgG antibodies over the first few weeks and after the exposure at week 27 showed a steep increase in antibodies that was not seen in the control group. This supports the theory that a second exposure results in a faster and stronger response.

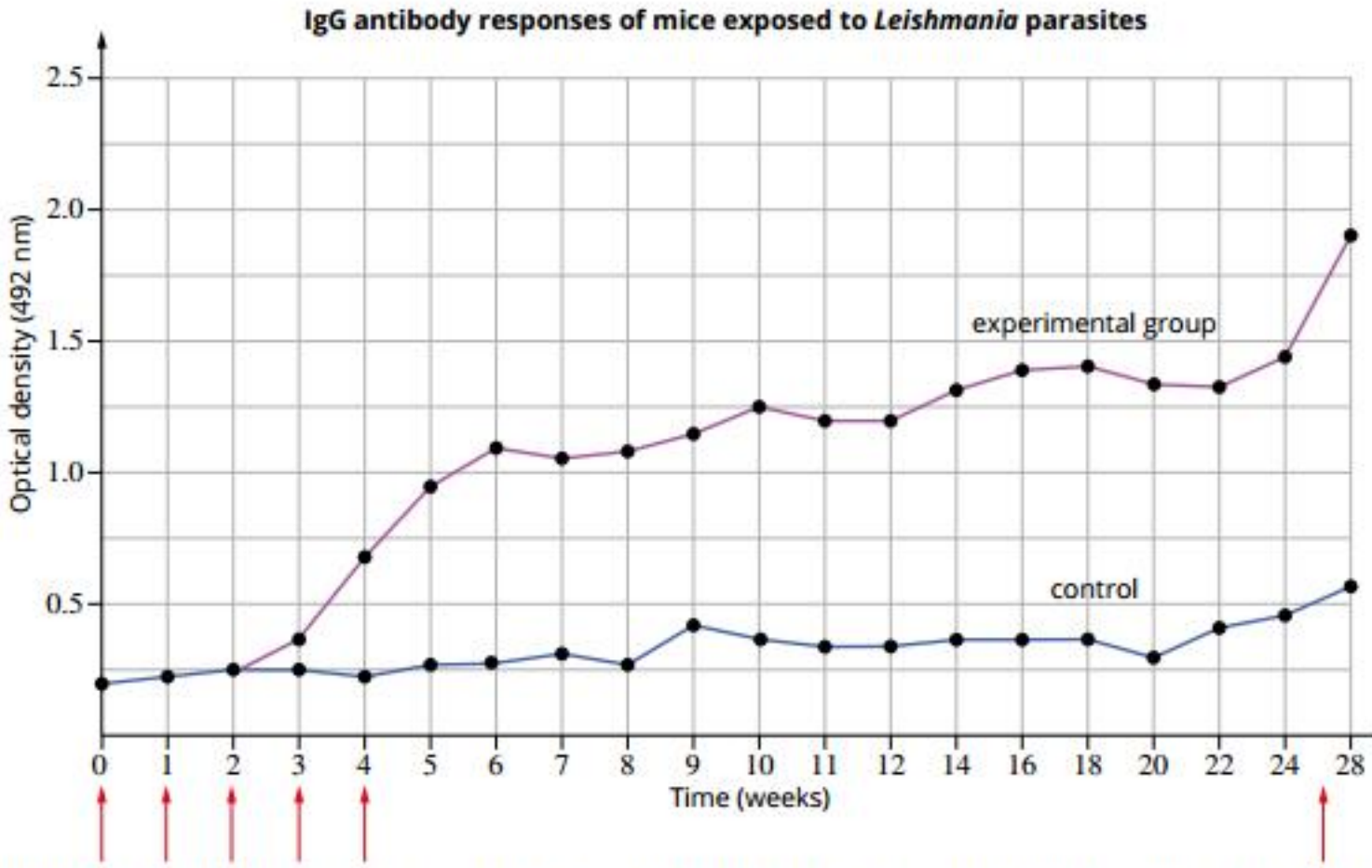


FIGURE 8.3.7 IgG antibody responses of mice exposed to *Leishmania* parasites (indicated by the arrows). Note that the graph for the experimentally bitten group (at 27 weeks) shows that a second exposure to an antigen produced a much quicker and stronger response than the initial exposure, which takes a long time to build up. Optical density is a measure of antibody concentration (Note: *x*-axis is not to scale.)

Artificial active immunity or vaccination

Active immunity is immune protection provided by an individual's own adaptive immune response. It can be primary (slow) or secondary (fast).

Artificial active immunity results from the administration of antigens to induce an adaptive immune response.

Preparations that artificially introduce the antigens of the pathogen to the immune system are called **vaccines**. The process of presenting a vaccine to the body is **vaccination** (also called **immunisation**). The antigens in the vaccine can be in the form of altered, weakened or killed pathogens, such as bacteria or viruses, or inactivated toxins or proteins produced by the pathogen.

Vaccines must be highly specific to initiate an adequate immune response that results in an immunological memory. Increased understanding of microbiology and immunology has led to the development of very safe vaccines that induce the desired immune response while producing minimal side effects.

There are different methods of producing vaccines.

Live attenuated vaccines

Live attenuated vaccines involve a living microbe that has been weakened in the laboratory, usually through repeated culturing. The advantage of live attenuated vaccines is that a single dose usually provides long-lasting immunity because the vaccines induce a strong adaptive immune response that produces many types of antibodies directed against multiple antigens.

The disadvantages of live attenuated vaccines are that although they are safe for most people, they may cause disease in people with weakened immune systems. Also, they may cross the placenta in pregnant women and damage the fetus.

Attenuated vaccines are more commonly used for viruses than for bacteria, because bacteria have thousands of genes and thus are much harder to control. Attenuated vaccines are used against measles, mumps, rubella and polio.

Inactivated vaccines

Inactivated vaccines, also known as **killed vaccines**, contain microbes that have been inactivated by heat, radiation or chemicals. The advantages of inactivated vaccines are that they result in the production of many different antibodies because they contain many different antigens. Inactivated vaccines can safely be used in people who have weakened immune systems.

The disadvantage of inactivated vaccines is that they stimulate a relatively weak immune response compared to live, attenuated vaccines, so they require booster doses to achieve and maintain long-term immunity. **Adjuvants** can be added to inactivated vaccines to help boost the immune response. Adjuvants are chemicals such as aluminium phosphate and aluminium hydroxide that stimulate a stronger immune response against antigens administered at the same time.

Most vaccines against bacteria are inactivated vaccines. Examples of inactivated vaccines are the inactivated rabies and hepatitis A vaccines.

Subunit vaccines

Like inactivated vaccines, **subunit vaccines** do not contain any live microbial components. Unlike inactivated whole-cell vaccines, subunit vaccines contain only parts of microbes selected for their ability to induce an adaptive immune response. Subunit vaccines can contain a fraction of an antigen, a single antigen or multiple antigens. These antigens can be proteins, detoxified toxins or polysaccharides. Subunit vaccines that contain multiple antigens induce a broader immunity, because they induce the production of antibodies directed against multiple antigens.

Subunit vaccines have the same the advantages as inactivated vaccines in that they are safer, more stable and easier to store than live, attenuated vaccines. However, subunit vaccines also share the disadvantages of inactivated vaccines in that that they require multiple doses and an adjuvant to improve the strength of the immune response.

Subunit vaccines are made by growing the pathogen in the laboratory and chemically extracting the antigens, or by using **recombinant DNA technology**. An example of a recombinant subunit vaccine is one that has been genetically engineered to produce a purified component of the protein coat of the virus that causes foot-and-mouth disease. Recombinant DNA technology can also be used for live vaccines, to genetically modify microbes so that they elicit an immune response but do not cause illness.

A **toxoid vaccine**, a type of non-recombinant subunit vaccine, uses toxins inactivated by formalin (called toxoids) to stimulate an adaptive immune response. Although the toxoid is inactivated, it remains similar enough to the original toxin that the immunological memory for the toxoid is also effective for the toxin. For example, *Clostridium tetani* is a bacterium that produces a neurotoxin that causes tetanus, and the inactivated toxin is used in the vaccine for tetanus. *Clostridium tetani* is illustrated in Figure 8.3.8. Another example of a toxoid vaccine is the diphtheria vaccine. Toxoid vaccines often require multiple doses to achieve immunity.

FIGURE 8.3.8 A digital illustration of *Clostridium tetani*, the bacterium that produces a toxin called tetanospasmin. The toxin acts on the central nervous system, causing muscle spasms that can result in convulsions, difficulty breathing and abnormal heart rhythms.

mRNA vaccines

A mRNA vaccine works by introducing a mRNA strand into the cells of the person to be vaccinated. This strand of mRNA is the code to make an important protein/antigen present on the pathogen. The mRNA is read by the ribosomes of the person's cells and the foreign antigen is made in abundance. These foreign antigens are then identified by the immune system and a primary immune response occurs.

Immunisation programs

Vaccination introduces the antigens from the pathogen to a person's immune system, generating an immune response where both antibodies and memory cells are formed. The reduced concentration of antigens presented in this way often means more than one challenge to the immune system is required in order to generate sufficient memory cells for protection from the wild pathogen. For this reason, the immunisation schedule for many diseases involves booster injections. These present the antigens to the immune system again.

The recommended vaccination schedule for Queensland is shown in Table 8.3.3 on page 348. Table 8.3.3 shows that vaccination programs do not commence until babies are at least 6 weeks old. This is because vaccination is unlikely to prove successful before that age. Maternal antibodies present in newborn infants inactivate the antigens in the vaccination before the infant's immune system can mount a response. Figure 8.3.9 shows the antibody levels for different types of antibodies in the fetus and infant. Note that fetuses can begin producing IgM antibodies by approximately 3 months gestation.

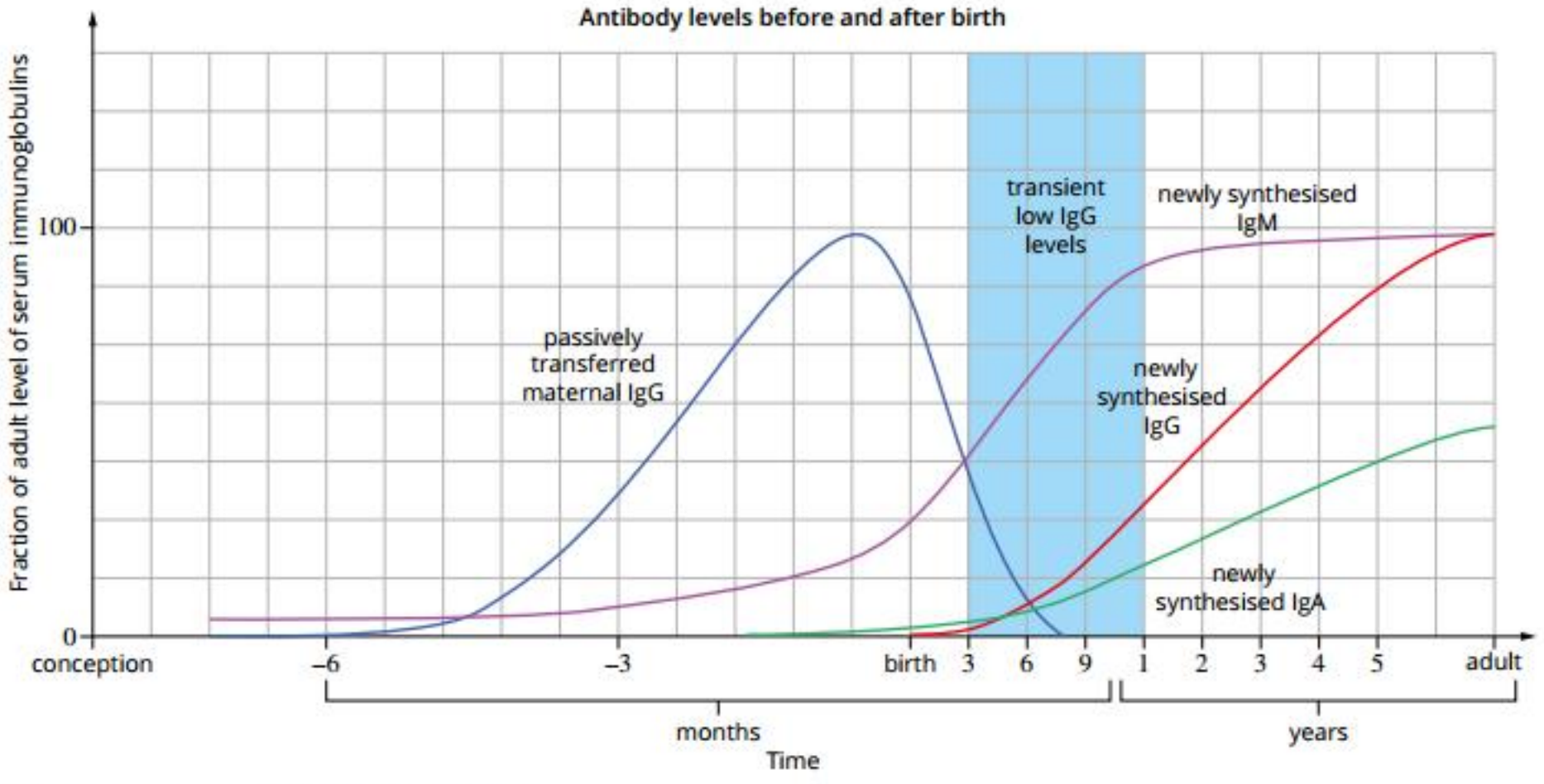


FIGURE 8.3.9 Antibody levels before and after birth

TABLE 8.3.3 Recommended vaccination schedule for Queensland

Recommended age for vaccination	Vaccines
Birth	• Hepatitis B • Respiratory Syncytial Virus (RSV)
6 weeks to 2 months	• DTPa-hepB-IPV-Hib • pneumococcal • rotavirus • meningococcal B
4 months	• DTPa-hepB-IPV-Hib • pneumococcal • rotavirus • meningococcal B
6 months	• DTPa-hepB-IPV-Hib • pneumococcal
6 months to 5 years annually	influenza
12 months	• measles, mumps, rubella (MMR) • meningococcal ACWY • meningococcal B • pneumococcal
18 months	• measles, mumps, rubella, chickenpox (varicella) (MMRV) • diphtheria, tetanus, pertussis, • *Haemophilus influenzae* type B
4 years	DTPa-IPV
Year 7 students	• DTPa-IPV • Human papillomavirus (HPV)
Year 10 students and 15–19 year olds	• meningococcal ACWY • meningococcal B
All adolescents	influenza
65+ years	• *Varicella zoster* (shingles) • influenza
70+ years	pneumococcal
All ages	influenza
Pregnant women from 20 weeks of pregnancy	Diphtheria, tetanus, pertussis
Also recommended for Aboriginal and Torres Strait Islanders	
Birth	tuberculosis
6 months	pneumococcal
18 months	hepatitis A
4 years	• pneumococcal • hepatitis A
50+ years	• pneumococcal • *Varicella zoster* (shingles)

DTPa-IPV = diphtheria, tetanus, pertussis, poliomyelitis.
DTPa-hepB-IPV-Hib = diphtheria, tetanus, pertussis, hepatitis B, poliomyelitis, *Haemophilus influenzae* type B.

Vaccine effectiveness

As can be seen from the vaccination schedule in Table 8.3.3, some vaccines, such as MMRV, are effective with one dose whereas others require multiple doses to maintain immunity. This is because the life span of the memory cells that produce the antibodies needed for active immunity is very variable.

For example, memory cells and antibodies against *Bordetella pertussis* (whooping cough) do not last throughout life. In a study in Thailand, it was shown that despite a significant infant vaccination program, covering 95% of all infants, by age 11 years nearly 50% of the population were antibody negative and at risk of developing *Bordetella pertussis* infection (Figure 8.3.10).

Unlike memory cells for whooping cough, influenza memory cells seem to be very long lived. In a recent study, survivors of the 1918 H1N1 influenza pandemic were tested for immunity to the virus, which had recently been isolated from preserved tissues. The individuals, now in their nineties, supplied blood samples, which were incubated with samples of the virus that caused the epidemic. The blood serum contained antibodies that bound tightly to the virus even though the survivors had not been exposed to it for over 80 years.

Influenza (or flu) viruses undergo antigenic drift relatively quickly. Antigenic drift is a change in the genes of the virus. When antigenic drift occurs, the antigens on the virus change. As antibodies are highly specific to their antigen, the immunity developed to the strains circulating in one year often won't provide protection against the newly evolved strains circulating in the population in future years. This is true whether you have caught the flu and developed natural active immunity or have received a flu vaccine and developed artificial active immunity.

To keep up with antigenic drift, new flu vaccines are released every year. Although it is a single injection, each new flu vaccine is a cocktail of vaccines for different influenza strains. The decision on which strains to vaccinate against is based on what strains are predicted to be the most common in the coming flu season. However, sometimes the most common strains are not the expected ones, in which case the flu vaccine provides little protection. This is most likely if the virus experiences antigenic shift, swapping genes with another strain of virus.

In 2009, a new H1N1 pandemic strain of influenza (Figure 8.3.11) infected humans. The 2009 strain was sufficiently different from the 1918 H1N1 strain that exposure to the earlier virus gave little protection. Minimal protection was seen in some patients who had had influenza vaccinations for a number of years, suggesting that some earlier versions of seasonal flu may have had some commonalities with the new strain.

Even though regular booster vaccines are needed for some diseases, the introduction of routine vaccinations has reduced the mortality and morbidity of disease in Australia and throughout the world.

Vaccination has had the greatest impact on mortality in developing countries. In economically developed countries, where nutrition is good and people can easily access high-quality healthcare, death rates from most infectious diseases were decreasing even before vaccination. Good hygiene and sanitation also significantly limited the spread of disease. For example, measles had a death rate of around one death per 100 cases before vaccinations began, and by 1990 had fallen to one death for every 2000 cases. However, in developing countries, where children are often malnourished and may be infected with other parasites, such as malaria and helminths (worms), mortality rates range between one in 200 and one in 17 under normal conditions to one in three in times of war or social disruption.

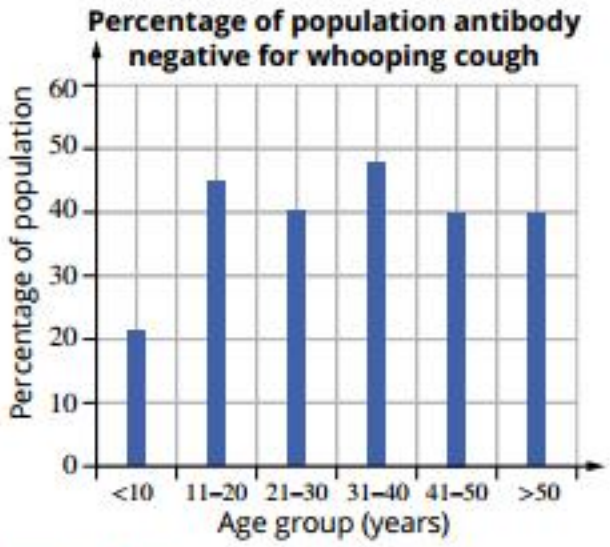


FIGURE 8.3.10 The percentage of Thai population with negative antibody titres for *Bordetella pertussis* (whooping cough)

Strains of influenza A virus are named according to their two surface proteins: haemagglutinin (H) protein and neuraminidase (N) protein. There are 18 haemagglutinin subtypes (H1–H18) and 9 neuraminidase subtypes (N1–N9).

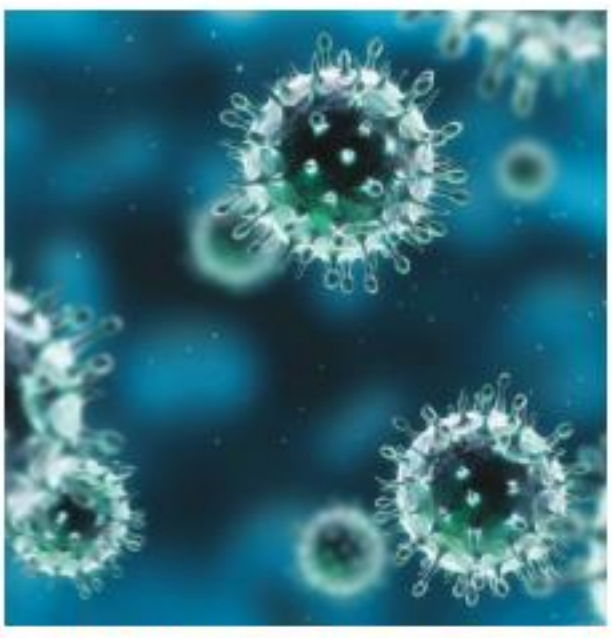


FIGURE 8.3.11 A digital illustration of the H1N1 strain of swine influenza virus

Morbidity is the state of being diseased or in ill health. Mortality is a measure of death rate.

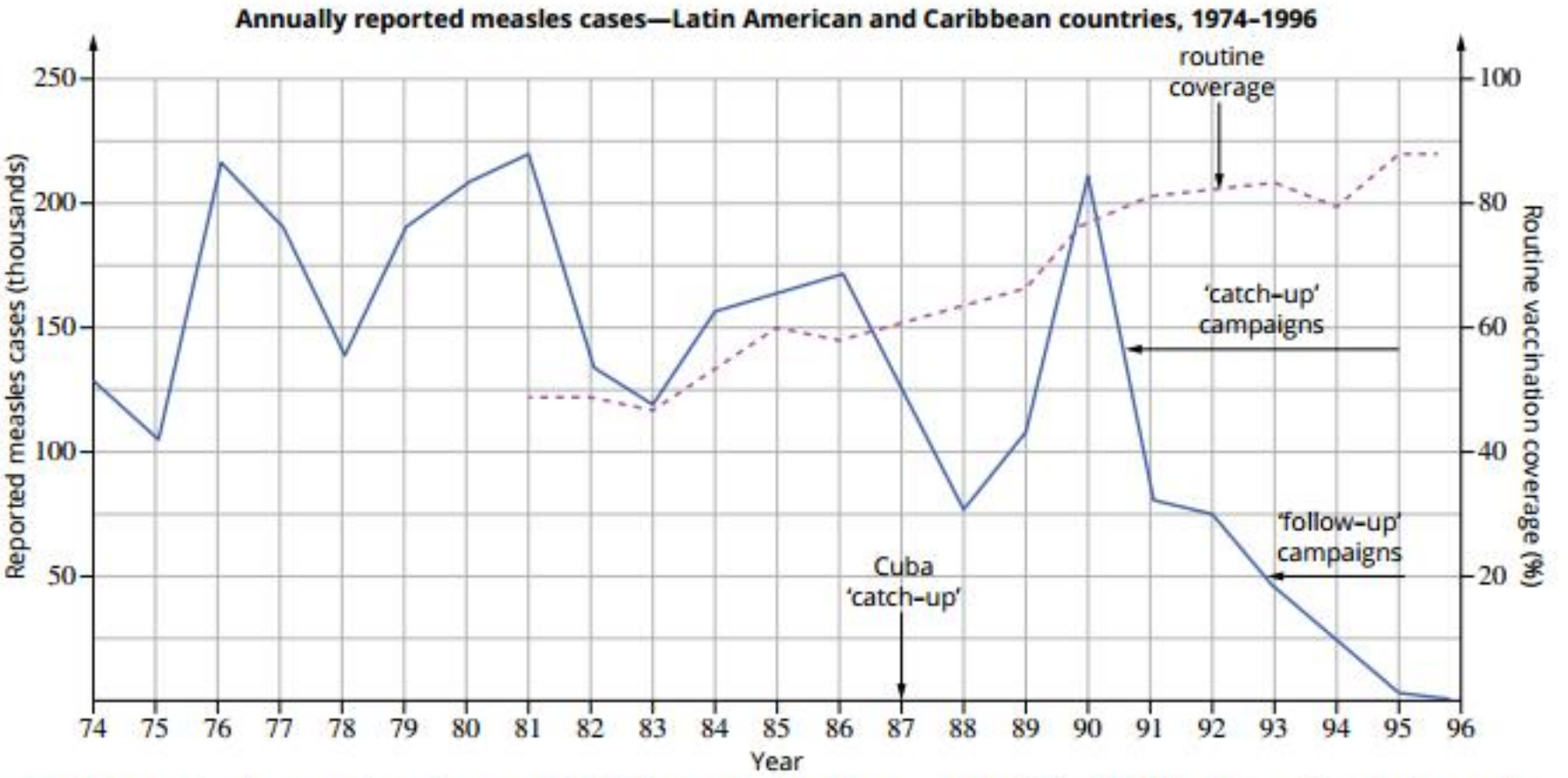


FIGURE 8.3.12 Annually reported measles cases in Latin American and Caribbean countries 1974–1996. The number of reported cases of measles in the Caribbean and Latin America decreased significantly as vaccination rates increased, indicating the effectiveness of vaccination programs.

FIGURE 8.3.13 This person is suffering from a mild case of smallpox. Note the many smallpox pustules that cause scarring as they heal. Most people would be covered in pustules over most of their body.

According to the World Health Organization, there were 134 200 deaths from measles in 2015. However, with about 85% of the world's children receiving at least one dose of measles vaccine, around 20.3 million deaths were prevented. The effect of vaccination on the incidence of measles in a developing area of the world, Latin America and the Caribbean, can be seen in Figure 8.3.12. Plummeting incidence rates mean the numbers of deaths also decrease even if rates per individuals infected do not improve.

The greatest success story for vaccination is smallpox. Smallpox was caused by the *Variola* virus. It was a terrible disease with a high mortality rate of around 30%. It was highly infectious and spread through droplets in the air. Those who survived were usually very disfigured by scarring left when the pox healed.

Smallpox virus was a good candidate for eradication by vaccination. It was a terribly feared disease that people would take almost any measure to avoid. It had a short incubation time, and the symptoms (pox or blister-like sores) were distinctive, so outbreaks could be quickly identified and contained. Figure 8.3.13 shows a person suffering from smallpox. Also, it was entirely confined to humans, which is essential if a disease is to be eradicated because if animals are a source of the disease, the only way to eradicate it is to eradicate the animal reservoir as well. A disease is eradicated when it has been removed from the world.

In 1980, the world was declared smallpox free (Figure 8.3.14). It had been eradicated. Today, the only source of *Variola* virus is in two high-security laboratories at the Centers for Disease Control and Prevention in Georgia, USA, and the State Research Centre of Virology and Biotechnology (VECTOR Institute) in Koltsovo, Russia.

The most recent disease that is being targeted for eradication is polio, which is caused by poliovirus. Eradication of this virus is close. As a result of a worldwide vaccination effort, numbers of people infected annually have fallen from 350 000 to 42 cases in 2016. Only three countries are known to have had cases of wild-type infection: Pakistan, Afghanistan and Nigeria. Each of these three countries is currently in a state of internal turmoil, which has disrupted vaccination programs.

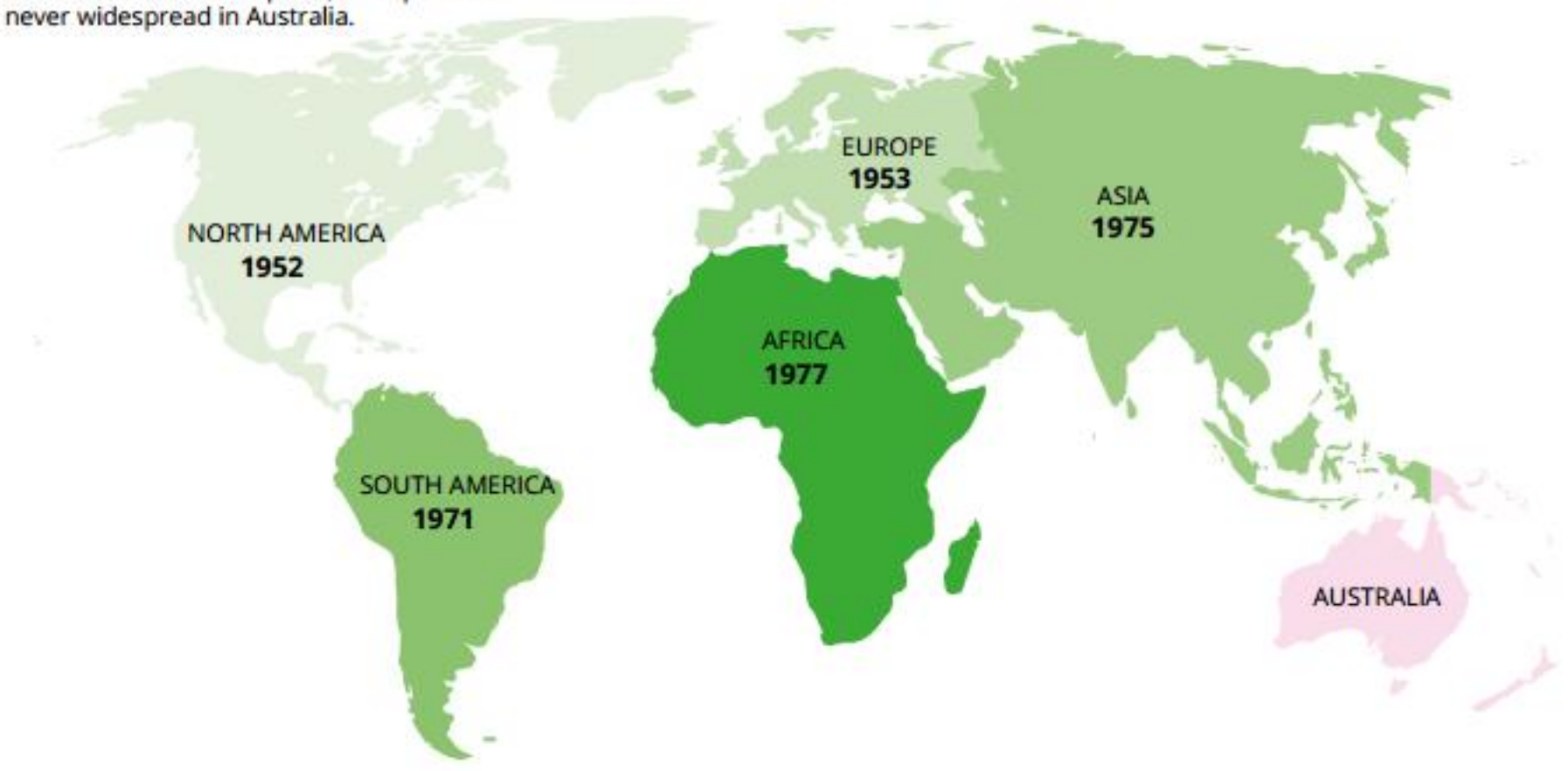


FIGURE 8.3.14 The timeline for the eradication of smallpox in the world. It was finally declared eradicated by the World Health Organization (WHO) in 1980, 3 years after the last reported case.

Herd immunity

Immunisation is critical, not only for the person immunised, but also for the wider community. For an immunisation program to be successful, a sufficient number of people need to be vaccinated; this is called **herd immunity** and is shown in Figure 8.3.15 on page 352. The more people who are vaccinated, the less chance there is of an infectious agent spreading throughout a population, because there are fewer potential carriers. Herd immunity is essential for protecting people who cannot be vaccinated or who have suppressed immune systems. This includes newborn babies, pregnant women, the elderly, people suffering from an immune disease and people taking immunosuppressant medication.

Breakdown of herd immunity: whooping cough

Immunological memory for some pathogens reduces over time, thereby reducing the herd immunity of immunised populations. An example of the breakdown in herd immunity in Australia is the recent spike in cases of whooping cough, a disease caused by the bacterium *Bordetella pertussis*. Although it only causes a persistent cough in adults, approximately one in 200 babies under the age of 6 months who become infected die. Babies cannot be vaccinated until they are 6 weeks old, and they are not fully protected by this vaccine until about 6 months of age.

One of the reasons for this breakdown in herd immunity is that not enough people get booster vaccinations. A public education campaign has been implemented to encourage adults to receive a booster vaccination to maintain herd immunity against whooping cough. New parents are offered the booster vaccination when their baby is born and are encouraged to recommend the vaccination to family and friends who will be in close contact with their baby.

a
No one is immunised, so infectious disease spreads easily through the population.
b
Some people are immunised, but infectious disease still spreads through most of the population.
c
Most people are immunised, limiting the spread of infectious disease in the population.
= not immunised but still healthy
= immunised and healthy
= not immunised, sick and contagious
= zone of contact with contagious person

FIGURE 8.3.15 The effectiveness of herd immunity. (a) With no immunisation in a community, infectious diseases spread easily. (b) With some immunisation in a community, infectious diseases spread less easily. (c) When most of the community is immunised, there are few carriers or infected people and minimal spread of infectious diseases. This is known as herd immunity.

In Australia, vaccination rates are among the best in the world but even here the rates fall short of the 95% that is considered necessary to fully provide herd immunity. As can be seen in Table 8.3.4, even Tasmania, which has the highest percentage of fully immunised children, has still not reached the target of 95% although rates of immunisation by age five have increased since 2005. Failure to achieve the targets leads to outbreaks of disease, such as the measles outbreak that occurred in Western Sydney in 2016.

TABLE 8.3.4 Rates of full immunisation of children by age 5 years in Australia by state from 2005 to 2017

State/territory	2005	2006	2007	2008	2009	2010	2017
Australian Capital Territory	78.42	83.53	87.57	86.17	85.32	89.41	93.52
New South Wales	71.89	78.72	83.74	76.58	80.61	87.60	93.48
Victoria	76.74	80.98	85.65	83.11	85.48	90.07	93.77
Queensland	76.93	82.29	83.83	81.24	82.22	88.22	93.48
South Australia	72.60	76.50	80.47	74.38	77.48	85.61	93.39
Western Australia	72.48	77.40	80.31	77.77	79.75	85.36	91.45
Tasmania	75.15	80.50	83.89	80.60	82.92	90.07	94.06
Northern Territory	75.95	78.80	83.61	81.40	81.80	85.77	92.39
Australia	**74.42**	**79.83**	**83.71**	**79.39**	**81.97**	**88.02**	**93.32**

Throughout Australia, immunisation rates of children (by 5 years of age) vary by area from the protective rates required to achieve herd immunity. Western New South Wales and Murrumbidgee, NSW, have protective rates that achieve herd immunity at 96.1% and 96.0%, respectively. The protective rates in North Coast, NSW (90.3%), Perth North, Western Australia (90.6%), Perth South, WA (90.8%), and Gold Coast, Queensland (90.8%), have concerning rates that are below what is required to achieve herd immunity.

Antimicrobials

Most of the time immune systems deal effectively with pathogenic invaders. However, sometimes the pathogen reproduces too quickly, and the immune system is unable to respond rapidly, which leads to serious illness or even death. In this case doctors today have a whole range of chemicals that they can use to reduce the ability of pathogens to reproduce or to kill them. These chemicals are known as antibiotics.

Many of the most common antimicrobials used to treat infection were originally found in plants or fungi. Some of the new formulations are modifications of the original plant or fungus derived substance. The first of these to be isolated and used was penicillin, which came from a fungus, *Penicillium rubens*.

Antibiotics work in a variety of ways. They may be bactericidal and kill the bacteria or bacteriostatic and stop bacterial reproduction. Table 8.3.5 below summarises the types of antibiotics in use and their effects of bacteria.

TABLE 8.3.5 The classes of antibiotics in use today and what they do

Action		Classes of antibiotics	Examples
bactericidal	inhibits synthesis of bacteria cell walls	β-lactams	penicillin amoxicillin
		glycopeptides	vancomycin
	inhibits the synthesis of bacterial proteins	aminoglycosides	streptomycin
		streptogramins	pristinamycin
	inhibit the synthesis of RNA by bacteria	annamycin	rifamycin
	inhibit DNA replication and transcription in bacteria	quinolones	ciprofloxacin
	disrupt bacterial cell membranes	lipopeptides	surfactin
bacteriostatic	prevents bacterial growth and multiplication	sulfonamides	sulfadiazine
	inhibits bacterial protein synthesis	chloramphenicol	chloramphenicol
		tetracyclines	tetracycline doxycycline
		macrolides	erythromycin
		oxazolidinones	cycloserine

8.3 Review

SUMMARY

- Immunity is active and passive.
- Naturally acquired active immunity results from surviving an infection by a pathogen.
- Artificially acquired active immunity arises as the result of the introduction of specific antigens into the body, usually by injection.
- A vaccine is a preparation that contains antigens from a pathogen and which induces active immunity in the recipient.
- mRNA vaccines induce the host to make the antigens to which the host's immune system reacts.
- Vaccines can contain live attenuated pathogens, killed pathogens, antigens from pathogens or toxoids produced by pathogens.
- Vaccines provide effective protection from disease, giving protection against influenza and are responsible for the eradication of smallpox.
- Artificial passive immunity involves the administration of a serum containing antibodies made in another organism.
- Natural passive immunity occurs when antibodies pass from mother to baby across the placenta or in breast milk.
- Haemolytic disease of the newborn occurs when the mother has made antibodies against the Rh antigen on the fetal blood and these antibodies cross the placenta during pregnancy.
- Herd immunity occurs when a sufficiently large proportion of a population has immunity to a specific pathogen so that the chances of a non-immune individual coming in contact with an infectious individual is remote.

KEY QUESTIONS

Describe

1 **a** Define 'passive immunity'.
b Describe the two ways in which passive immunity is produced.

2 Identify which type(s) of immunity provide long-term protection from disease.

3 Explain the function of adjuvants. Include a definition of adjuvant in your answer.

4 What causes haemolytic disease of newborns?

Apply

5 Distinguish between eradication and elimination in terms of disease.

6 Explain why smallpox was a good candidate for disease eradication.

7 Compare artificial and natural passive immunity.

8 Compare artificial and natural active immunity.

9 Under which circumstances is passive immunity artificially induced?

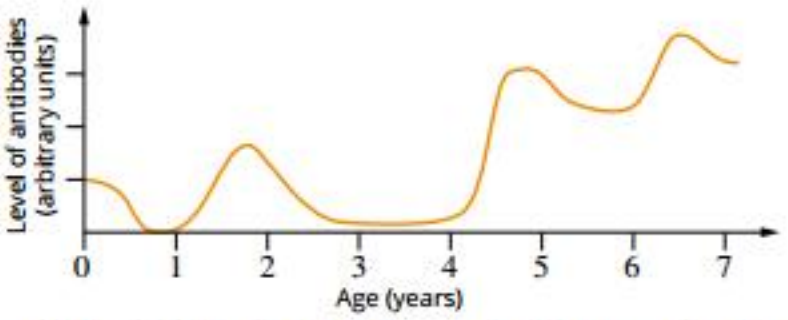
10 Explain why many vaccinations need a series of doses in order to establish and maintain immunity.

11 Live attenuated vaccines tend to give longer lasting protection than subunit vaccines. Explain why this is the case.

12 **a** Queensland Health Authorities recommend that pregnant women should be vaccinated against tetanus, diphtheria and pertussis from 20 weeks of pregnancy onwards. Suggest why is this recommendation given.
b It is also recommended that ideally the vaccination should be given before the 33rd week of pregnancy. Suggest reasons for this.

Analyse

13 The following graph shows the levels of measles antibodies in a child from birth to 7 years of age.



a This child had antibodies against the measles virus at birth. Explain how.

b Explain why the antibody levels dropped to zero in the months following the birth.

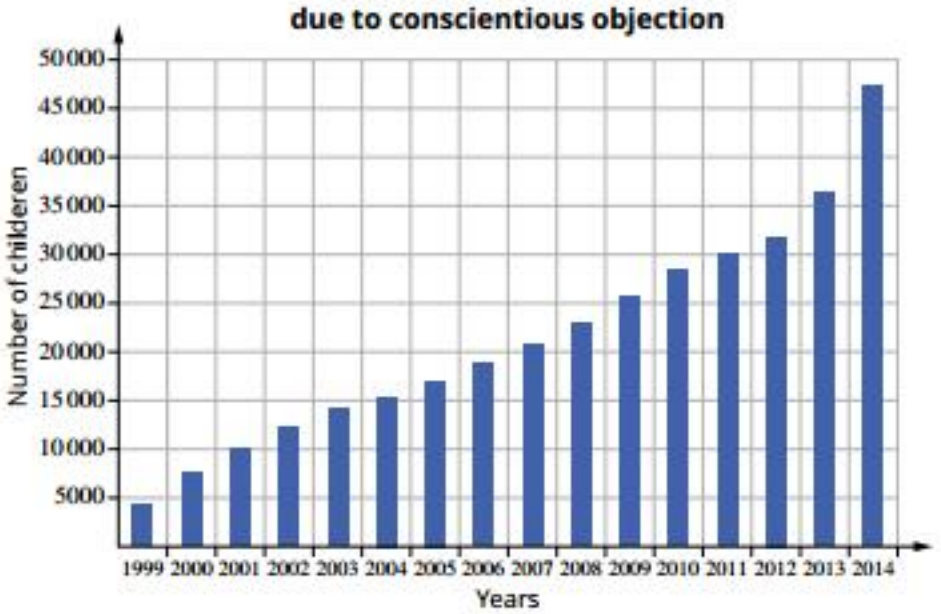
c The child was immunised against measles at 1 year of age and again at 4 years of age.

- **i** Assess with conceptual justification the results of the 1-year and 4-year immunisations.
- **ii** Draw a conclusion about the level of antibodies between the ages of 3 and 4 years.

d At 6 years of age, antibody production increases greatly again, but no vaccination had occurred. Infer an explanation.

e In Australia, the number of measles infections in children has been increasing recently, with a jump in 2015–16. Argue why, using the data in the following graph.

Australian children not immunised due to conscientious objection



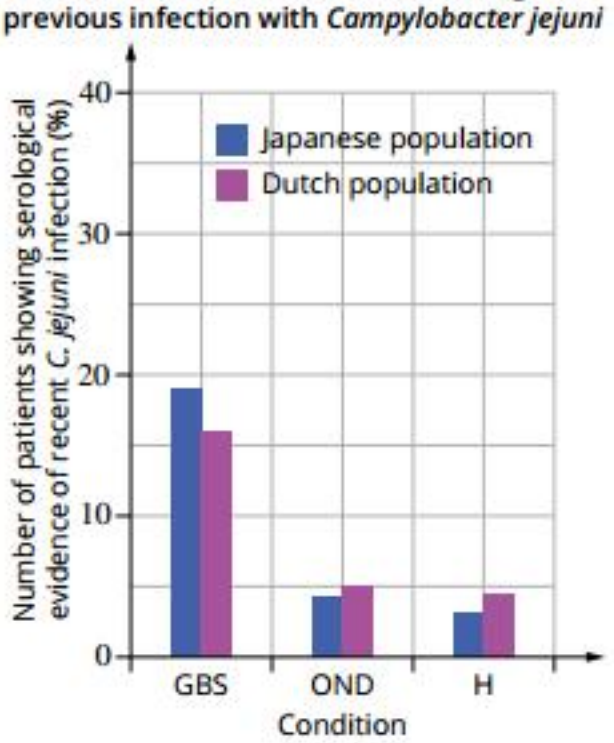
14 Guillain–Barré syndrome (GBS) is a neurological disease. It is characterised by paralysis, which occurs when an autoimmune reaction causes damage to nerves by the patient's own immune system. GBS generally follows a previous infection. It is not confined to any particular sex or age group.

It has been suspected for sometime that one trigger for the development of GBS is infection with the bacterium *Campylobacter jejuni*. *C. jejuni* is a common cause of food poisoning. Some people can be infected and remain asymptomatic.

Some researchers have suggested that previous infection with *C. jejuni* is more likely to result in the development of GBS among Asian populations than among European populations. A study was undertaken to investigate this proposal. Patients from The Netherlands (Dutch) and Japan with GBS were assessed by ELISA assays to determine whether they had been infected with *C. jejuni* previously. A group having other neurologically based diseases (OND) was included, as were a healthy group (H).

The Japanese subjects were 88 with GBS, 27 OND and 56 H and the Dutch subjects consisted of 132 GBS, 42 ONO and 30 H. The results are shown in the following graph.

Patients with and without GBS having had previous infection with *Campylobacter jejuni*



a Propose reasons for including the OND and H groups in the study.

b Draw a conclusion regarding the hypothesis, 'People with Asian ethnicity are more prone to GBS following *C. jejuni* infection'.

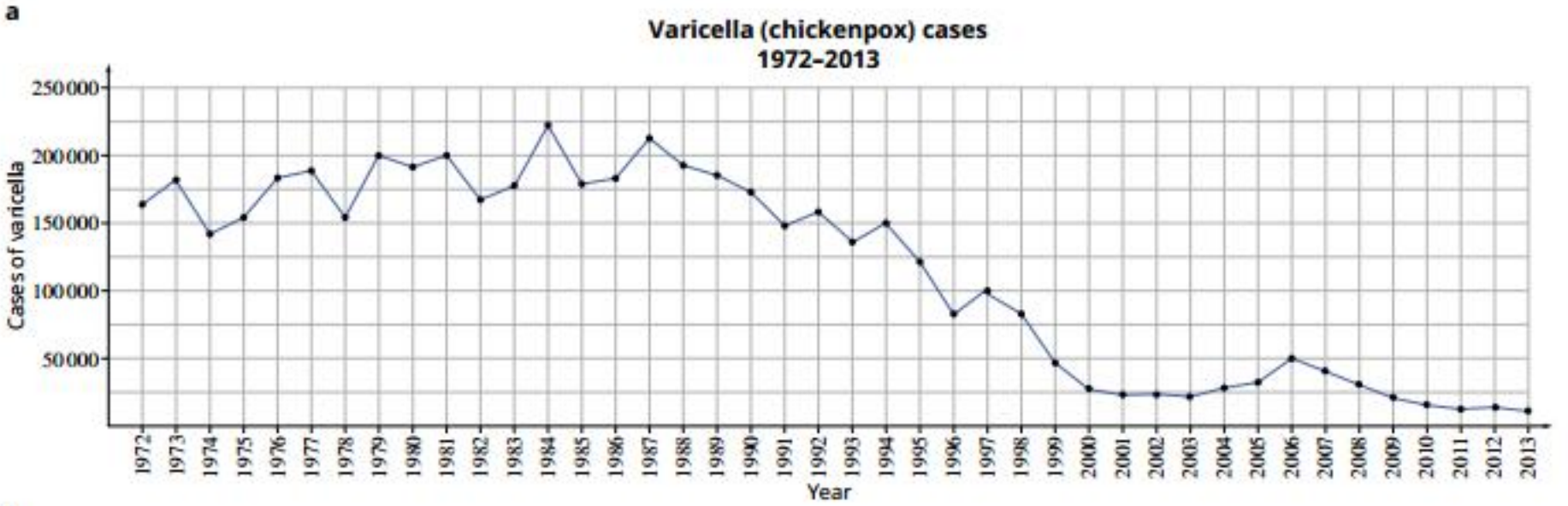
continued over page

8.3 Review *continued*

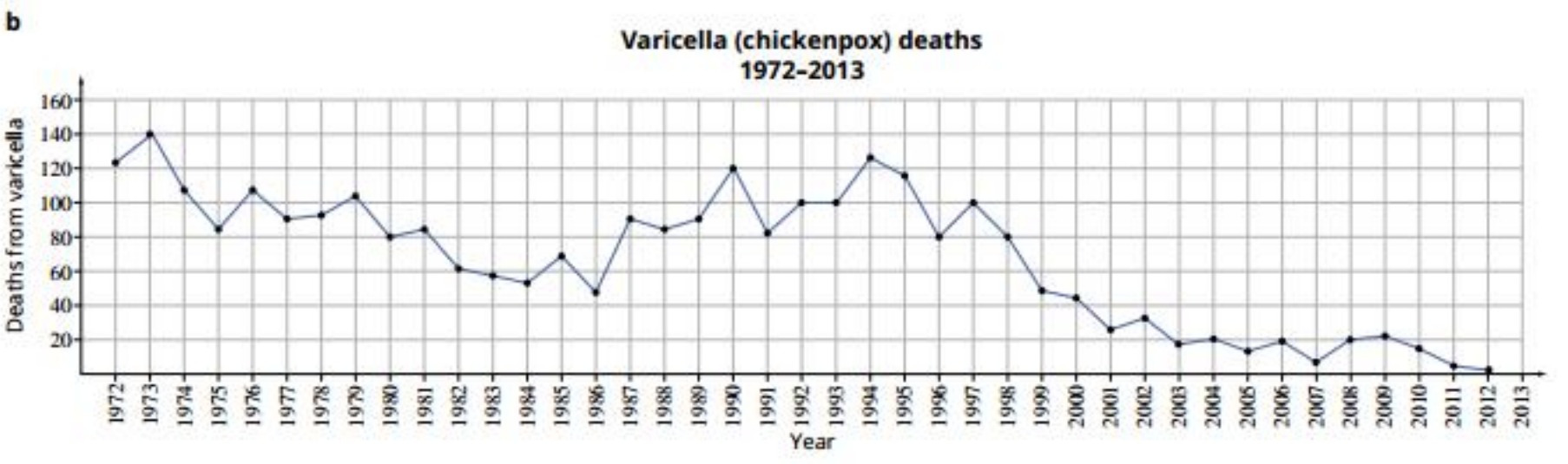
15 Consider the two graphs below for incidence (a) and mortality (b) from chickenpox (varicella) in the USA. The varicella vaccine was introduced in 1995. That year, there were 120 624 cases of varicella, and 115 deaths from varicella.

- **a** Determine the death rate per 1000 cases of varicella in 1995.
- **b** Estimate the average number of varicella cases per year before the introduction of vaccinations.

a



b



Chapter review

08

KEY TERMS

active immunity
adaptive immune response
adjuvant
agglutination
alkaloid
allergen
antigen–antibody complex
antigen–presenting cell (APC)
antiserum
apoptosis
artificial active immunity
artificial passive immunity
B lymphocyte (B cell)
cell-mediated immunity
chemokine
clonal expansion
clonal selection
complement protein
complement system
constant region
cyanogenic glycoside
cytokine
cytotoxic T lymphocyte (T_c)
defensin
dendritic cell
digestive enzyme inhibitor
epitope
fever
granzyme
haemolytic disease of the newborn
heavy chain
helper T lymphocyte (T_H)
herd immunity
histamine
human leukocyte antigen (HLA)
humoral immunity
hydrolytic enzyme
immunisation
immunogen
immunoglobulin (Ig)
inactivated vaccine
inflammation
innate immune response
interferon
interleukin
killed vaccine
leukocyte
light chain
live attenuated vaccine
lymphocyte
macrophage
major histocompatibility complex (MHC)
mast cell
megakaryocyte
memory B lymphocyte
memory T lymphocyte
microbe-associated molecular pattern (MAMP)
natural active immunity
natural passive immunity
passive immunity
pathogen-associated molecular pattern (PAMP)
pattern recognition receptor (PRR)
perforin
phagocyte
phagolysosome
phagosome
phenolic
plasma cell
platelet
precipitation
primary immune response
protease inhibitor
recombinant DNA technology
regulatory T lymphocyte
saponin
secondary immune response
self-tolerance
serum
subunit vaccine
T cell receptor (TCR)
T lymphocyte (or T cell)
terpene
toxoid vaccine
vaccine
vaccination
variable region
vasodilation

KEY QUESTIONS

Describe

1 Identify the chemical that is not produced by plants in response to infection by a pathogen.
   A saponins
   B lignins
   C terpenes
   D phenolics

2 Classify the type of immunity provided by vaccination.
   A artificial passive
   B natural active
   C artificial active
   D natural passive

3 Identify the statement that can most accurately be applied to the secondary immune response.
   A A small amount of antibody would be found in the blood serum of the patient.
   B It requires a large dose of the antigen in order to provoke an immune response.
   C It occurs with a time lag that is shorter than the primary response.
   D It results in large numbers of IgD molecules.

4 Identify the type of antibody found in the highest concentration in the blood of a newborn baby.
   A IgA
   B IgG
   C IgE
   D IgD

5 Identify which of the following must be present on an antigen-presenting cell membrane, in order to activate a T_H cell.
- **A** IgM
- **B** MHC-I markers
- **C** MHC-II markers
- **D** IgG antibodies

6 List three barriers to infection used by:
- **a** plants
- **b** animals.

7 Identify the cells that display MHC-I markers.

8 Define 'self-tolerance' and describe how it is achieved.

9 Which cells carry MHC-II markers?

10 Write the correct word or term for the definition given.
- **a** A cell that uses MHC-II on its surface to present foreign antigens to helper T lymphocytes.
- **b** The process of a particular B lymphocyte dividing rapidly to form many plasma cells.
- **c** The part of an antigen that binds to the antigen-binding site of a specific antibody.
- **d** A white blood cell formed or matured in lymphatic tissue.
- **e** A preparation of altered, weakened or killed microorganisms, or inactivated forms of toxins or antigens introduced into the body to stimulate active immunity.

11 Name one chemical defence possessed by plants and state how it helps.

12 Identify the cells that are usually the initial stimulators of the inflammatory response.

13 State two functions of the complement system in mammals.

Apply

14 What are TLRs and PAMPs? Explain how they are related.

15 Explain the role of inflammation in the immune response.

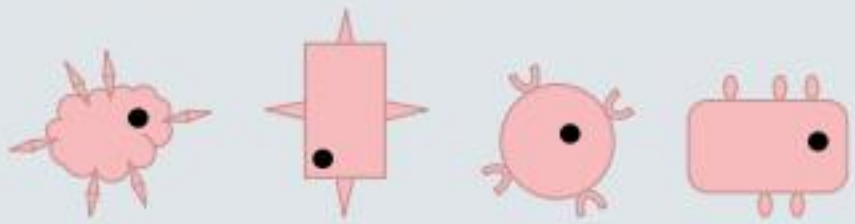
16 Compare macrophages and neutrophils

17 In a particular community, avian influenza strain H3N2 and a human strain H1N1 have been circulating. Without any warning, there is a sudden increase in people arriving at hospital emergency departments with influenza. The strain is analysed and found to be a new strain H3N1. This new strain is likely to have arisen as a result of:
- **A** lack of people in the population having vaccination
- **B** antigenic shift
- **C** antigenic drift
- **D** the development of antibiotic resistance by the influenza virus

18 Lymphocytes recognise cells as non-self because the foreign cells have antigens that are complementary in shape to their receptors for detecting foreign antigens.
- **a** Explain what is meant by the word 'complementary' in this context.
  A lymphocyte (cell Z) with its receptor is shown below.

cell Z

- **b** Explain which of the cells, A, B, C or D, will be identified as non-self by the lymphocyte.



cell A cell B cell C cell D

19 Cassava provides a significant proportion of the daily kilojoule intake for over 600 million people throughout the world. In both Africa and India, cassava crops are regularly attacked by the cassava mosaic virus (CMV). One important method of protecting crop production is to use cassava varieties that show a natural resistance to CMV. Propose one feature the CMV-resistant varieties of cassava plant could have that non-resistant plants do not.

20 Upon first exposure to an antigen, there is a lag time before antibody production. Explain why this occurs.

21 Chédiak–Higashi syndrome is a rare inherited disorder of the immune system in which the proteins that regulate the joining of lysosomes with endosomes in phagocytes are defective.
- **a** Name the defective structure.
- **b** The failure of lysosomal breakdown of engulfed bacteria seriously undermines not only the innate immune response, but also the adaptive immune response. Explain why.

22 On occasion, the blood bank in Melbourne advertises for donors who have recently recovered from chickenpox. The blood bank takes a blood donation from these people and then separates the blood by a centrifuge into the blood serum and blood cells. The blood cells may be injected back into the donor. The serum is then purified and some of the proteins are extracted.

- **a** Name these proteins.
- **b** These proteins are given to patients. Identify the group of patients that is most likely to be in need of these proteins.
- **c** Explain why the extraction of these proteins does not affect the donors' future immune response to chickenpox exposure.
- **d** Explain whether the injection of these proteins gives the recipients long-term immunity.

23 In a family of two children, child X, the elder, receives the whooping cough vaccination whereas the younger, child Y, does not. Child Y contracts pertussis (whooping cough) 2 years later and recovers in hospital and now both children have developed immunity against pertussis.

- **a** Compare the response of the B and T lymphocytes and antibodies when child Y contracts pertussis.
- **b** Considering that both children develop an immunity to pertussis, explain why it is advisable for children to be vaccinated.
- **c** Child Z from another family is currently undergoing chemotherapy and is immune-suppressed as a result. Discuss why her parents are concerned about some children not being vaccinated against pertussis.

Analyse

24 DiGeorge syndrome is a rare genetic disease in which a gene necessary for the proper formation of the thymus gland is missing. How might this affect the functioning of the immune system?

25 Saponins are a group of chemicals that defend plants against attack by pathogens. An investigation to examine one type of saponin, avenacin, was undertaken to investigate the research question, 'Will the absence of avenacin result in an increase in plant disease?'.

A group of plants from the species *Avena strigosa* were mutated so they could not produce avenacin. They were allowed to mature and were then exposed to spores of the pathogenic take-all fungus (*Gaeumannomyces graminis*).

The results of one study with the mutant plants are shown in the following table. Plants were examined 21 days after infection with the fungal spores. Wild-type plants do not have reduced avenacin production. Plants marked by * have reduced avenacin levels and other mutants lack any avenacin. Each group contained a large number of genetically similar plants.

Draw conclusions about the effectiveness of avenacin in combating fungal infections in plants.

Plant group	Disease rating, % seedlings			
	No disease	Low disease	Moderate disease	Severe disease
Wild type (not mutant)	100	0	0	0
A	0	25	12	63
B	28	28	28	16
C*	62	38	0	0
D	31	44	6	19
E	12	71	17	0
F	27	40	33	0
G	6	44	50	0
H*	56	38	6	0
I*	74	21	5	0

26 A study was undertaken to investigate the effectiveness of an extract from the herb *Gentiana cruciata* on the growth of four species of bacteria. 0.6 mL of extract of root containing 1.8 ± 0.06% tannins and 8.5 ± 0.09% polyphenols from the herb was placed in a central well in agar plates spread with bacteria and then incubated. After 24 hours, the zones of inhibition around the wells were measured and tabulated as seen below.

Bacterial species tested	Diameter of zone of inhibition (mm)
Staphylococcus aureus	25.2
Bacillus subtilis	22.0
Escherichia coli	20.1
Candida albicans	17.9

- **a** What do the results suggest about the effectiveness of *G. cruciata* extract as a bacterial inhibitor?
- **b** What addition to the experiment would have made the results more useful in determining its usefulness?

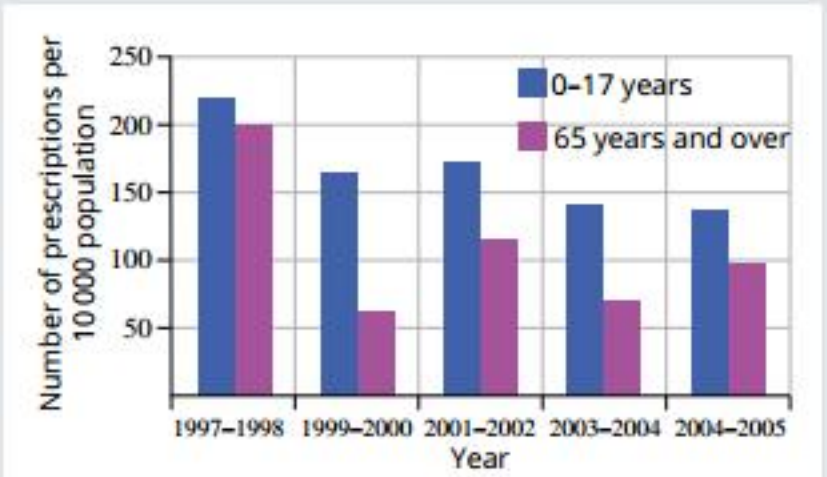
27 Chickenpox (varicella) is a disease caused by the *Herpes zoster* virus. Vaccination has been a part of the childhood immunisation schedule in Queensland since 2005. The vaccine is currently administered at 18 months of age.

- **a** Explain why 18-month-old children in Queensland do not contract the chickenpox disease from the vaccine containing a live virus.
- **b** Before the vaccination program, it was common for the eldest child in a family to develop chickenpox in their kindergarten or first year of school. After the eldest child's infection, the younger children would then develop the disease, but breastfeeding babies in the family rarely caught chickenpox and even if they did it was generally very mild. Explain this observation.
- **c** Since the introduction of chickenpox vaccine to the immunisation schedule, there has been an increase in the incidence of chickenpox among adults. Infer why this is occurring.

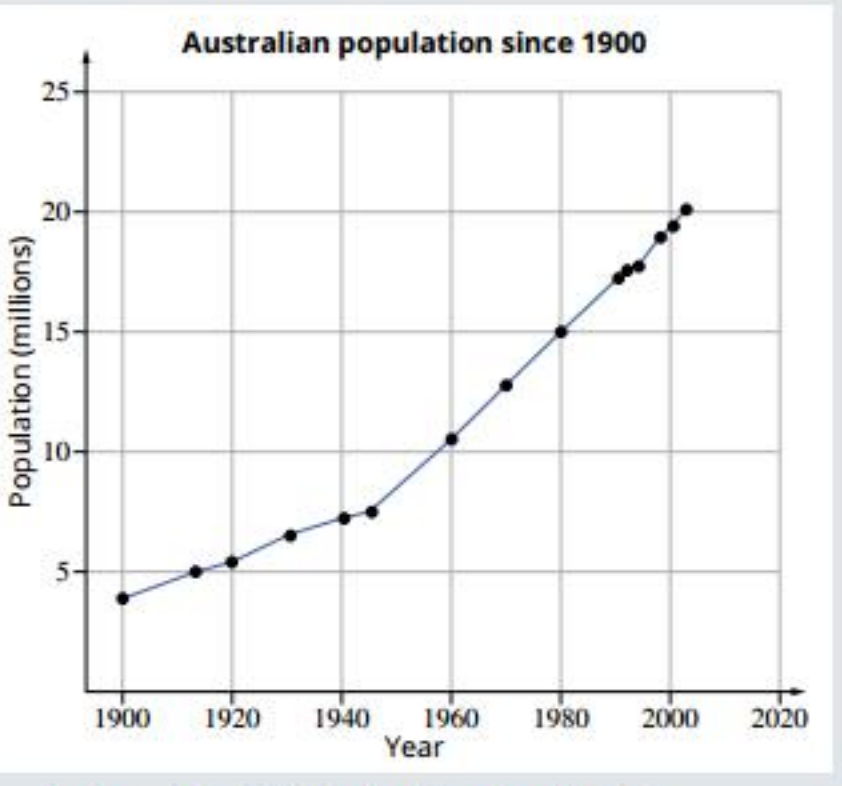
28 After World War I, a Spanish flu pandemic swept the world. More than 20 million people died. The Spanish flu was unusual because many of the fatalities occurred among young people with healthy immune systems. Modern medical opinion is that most of these deaths were caused by acute respiratory distress syndrome initiated by a 'cytokine storm'.
Cytokine storms occur as a result of positive feedback between macrophages and the cytokines.
Draw a flow chart to show how a cytokine storm can eventuate and cause death.

29 Research in the USA has shown that the common cold costs the US economy around $25 billion each year in lost productivity. The cost of the common cold to Australia would be proportionally similar. The major issue with the common cold is that there are many different strains of the rhinoviruses that cause colds.

- **a** The graph below shows the number of prescriptions for antibiotics issued in the USA as treatment for the common cold.



  - **i** Discuss the use of antibiotics for the treatment of the common cold.
  - **ii** Determine which group was most likely to receive a prescription for antibiotics to treat their cold, according to the data in the graph.
- **b** The following graph shows the population of Australia from 1900 to 2005.

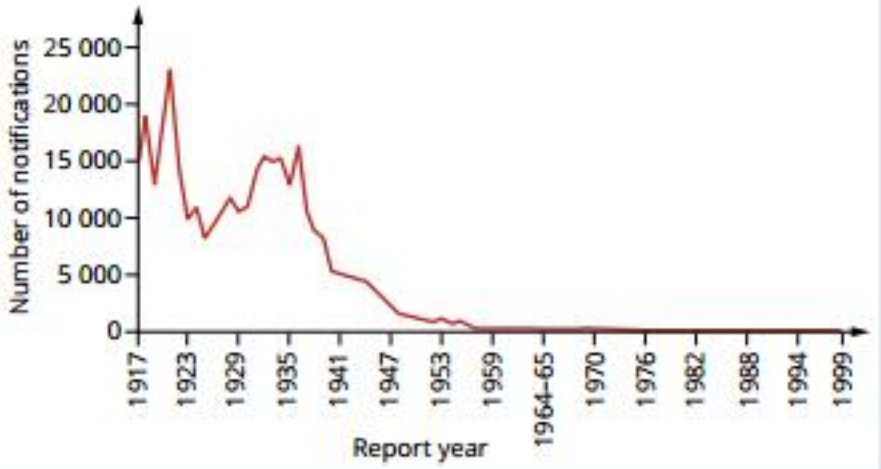


  - **i** Assuming that Australia's rate of issuing prescriptions for a cold is similar to that of the USA, use the information in the graphs to estimate how many people in Australia received a prescription for the common cold in 2000.
  - **ii** Each prescription for a course of antibiotics costs the Australian government approximately $20.00. Calculate how much money was wasted in 2000.

30 Diphtheria is a potentially fatal disease caused by either *Corynebacterium diphtheriae* or *C. ulcerans*. About 10% of infected individuals die. For this reason, diphtheria is one of the diseases for which children are vaccinated. The first vaccinations became available in 1921. By 1929, contacts of people with diphtheria were being vaccinated. School-based vaccination programs began in 1932.

Diphtheria is still a major health issue in some countries. Fewer than 10 cases have occurred in Australia in the last 10 years, all of which have been linked to overseas travel. The last recorded death from diphtheria in Australia was in 2011.

The graph shows the incidence of diphtheria in Australia between 1917 and 1999.



a Evaluate the effectiveness of the vaccination program in Australia.

b Explain why, despite the high incidence of diphtheria in some other countries and the ease of international travel, only isolated cases and no outbreaks of diphtheria have occurred in Australia in the last 10 years.

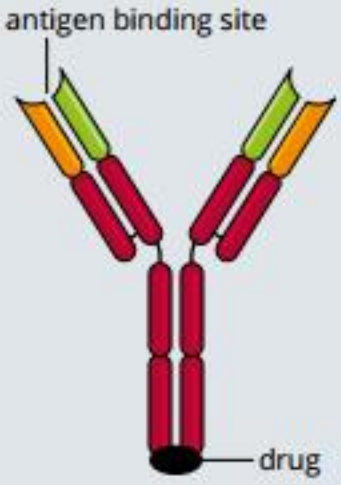
Interpret

31 Traditional cancer treatments have many side effects. This is because the drugs are highly toxic to cells and kill many healthy, non-cancerous cells as well as the cancer. Rapidly dividing cells such as hair follicles and the linings of the digestive tract are affected very badly.

Researchers have sought ways to make anticancer drugs more specific to cancer cells. Cancer cells have unique features that set them apart from other body cells, so scientists tried to exploit these differences to devise new treatments.

An important advance was the development of monoclonal antibodies. Monoclonal antibodies are artificially constructed antibodies with antigen–binding sites that allow them to attach to cancer cells with a much higher affinity than to normal body cells.

One group of researchers suspect that monoclonal antibodies can increase the efficacy and reduce the side effects of a commonly used anticancer drug by attaching the drug to a monoclonal antibody that has a binding site specific to an antigen that is expressed only on cancer cells.



The results of a preliminary trial comparing administration of the drug attached to the antibody with conventional administration are shown in the table below. Propose an extension of this experiment. Provide a rationale for your extension.

TABLE 1 Tumour response to antibody-boosted or conventional drug treatment

Patient number	Antibody or conventional therapy	Tumour size at start of treatment (mm^3)	Tumour size after one round of treatment (mm^3)	Change in tumour size (mm^3)	Change in tumour size (%)
1	antibody	12.5	9.6	−2.9	−23.2
2	antibody	23.9	15.5	−8.4	−35.1
3	antibody	54.2	26.8	−27.4	−50.6
4	antibody	46.8	27.9	−18.9	−40.4
5	antibody	53.6	56.4	+2.8	+5.2
6	conventional	54.8	48.5	−6.3	−11.5
7	conventional	84.1	66.9	−17.2	−20.5
8	conventional	36.9	30.8	−6.1	−16.5
9	conventional	56.1	49.1	−7.0	−12.5
10	conventional	38.9	31.5	−7.4	−19.0

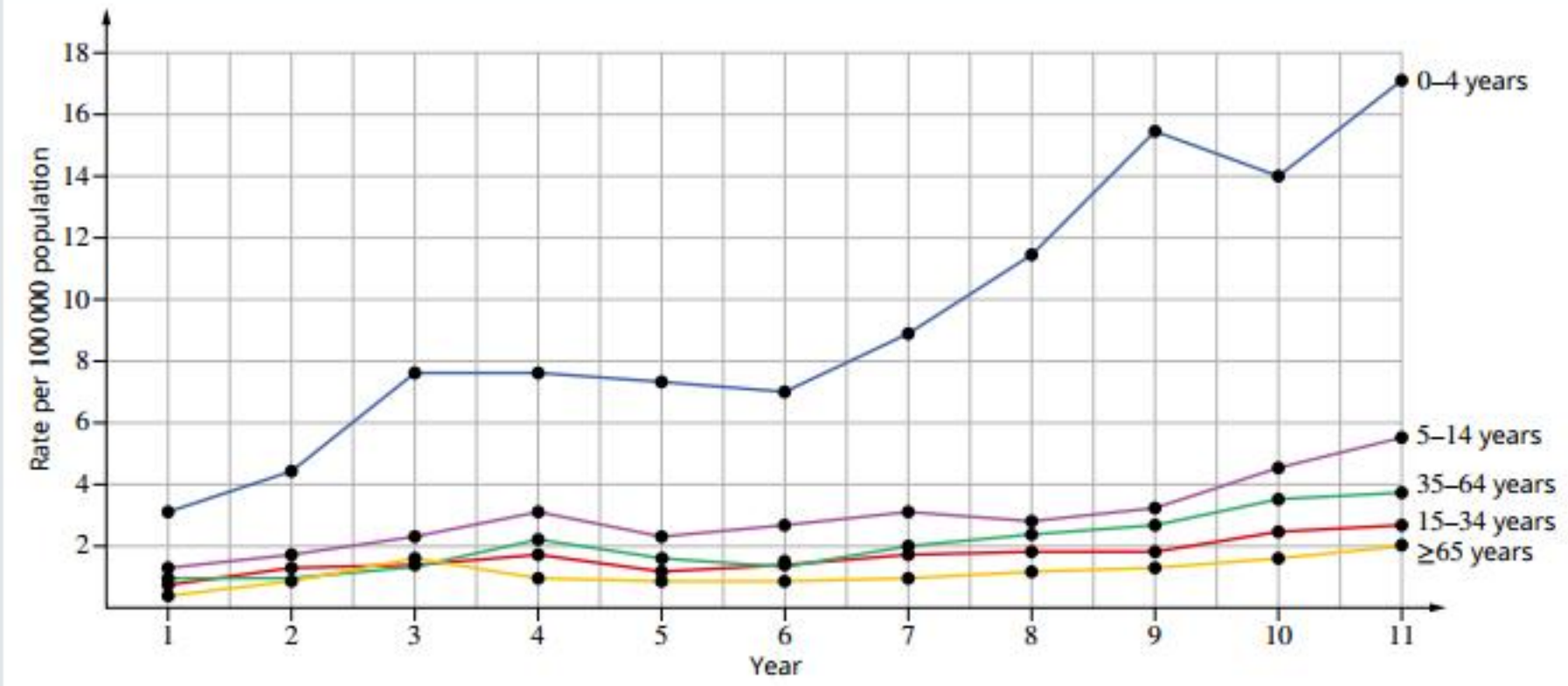
32 Allergic reactions are a life-threatening problem caused by the immune system. They occur when the immune system forms antibodies to antigens that do not pose a threat to the body. These antibodies attach to a type of immune system cell found in many tissues, called a mast cell. An allergic reaction occurs when the mast cell releases large amounts of histamine because the antigen (e.g. peanut or pollen) attaches to the antibodies on the cell. The production of large amounts of histamine can cause inflammation throughout the body, called anaphylaxis.

a Create a theoretical treatment for peanut allergy in Australia that is appropriate to the population. Provide a rationale with your proposed treatment.

b i Compare the approximate number of hospital admissions per 100000 in each age group in Year 11 to that in Year 1.

ii Assess whether all individuals maintain their allergic reactions throughout their life and propose a theoretical reason.

c A group of scientists think that they have developed a vaccine that would protect people at risk from developing allergic reactions to peanuts. Design an experiment, using mice that have a genetic tendency to peanut allergy, that would allow the scientists to test their new drug. State the results that would show that the vaccine is a success.



33 Japanese encephalitis virus (JEV) is a mosquito-transmitted disease. The most important vectors are mosquitoes in the genus *Culex*, especially *C. tritaeniorhynchus* and *C. vishnui*. JEV is of major public health importance in an area that extends from Japan to India.

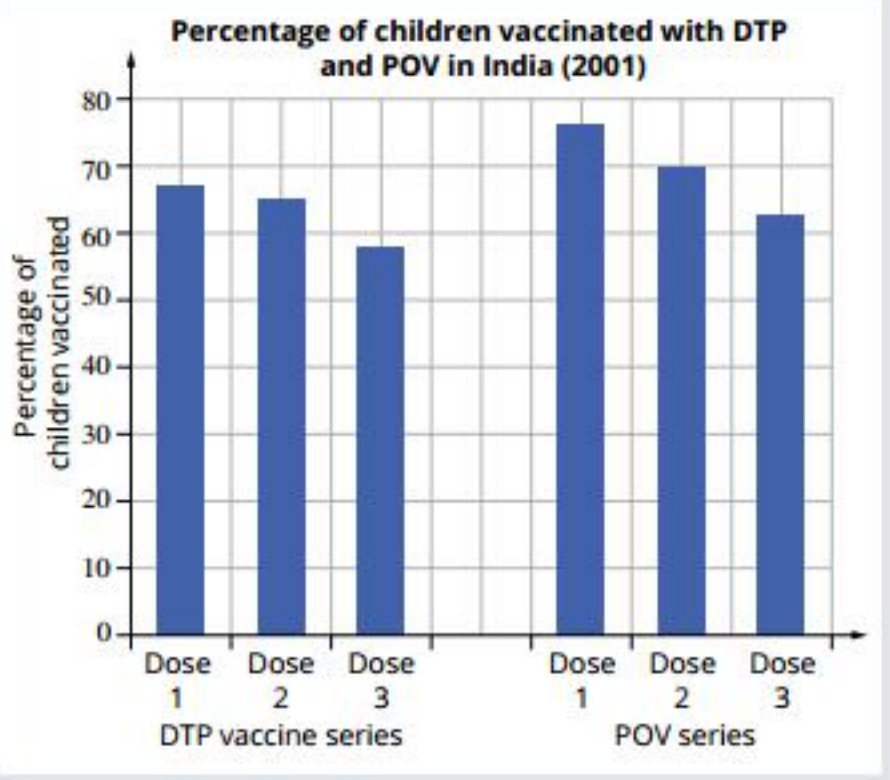
Most individuals do not suffer major symptoms but in a few cases encephalitis (brain inflammation) develops, and these patients have a 30% mortality rate. In one epidemic in 1924, 6000 people were infected, of whom 60% died. Children are at much higher risk of serious consequences, and fetal infection through the placenta from infected mothers has been observed. Severely affected individuals who survive may suffer brain damage, paralysis and/or deafness.

People in urban areas are less at risk because of the lower mosquito numbers and strategies that reduce the chances of being bitten, such as mosquito nets and insect repellents.

A government in eastern Asia is considering adding JEV vaccination to its childhood vaccination schedule. There is a vaccine available for JEV. It comes in two different forms. One form is given as a single dose and the other is given in three doses, two administered a week apart followed by a booster at one year. Both methods require a single booster dose at Year 3.

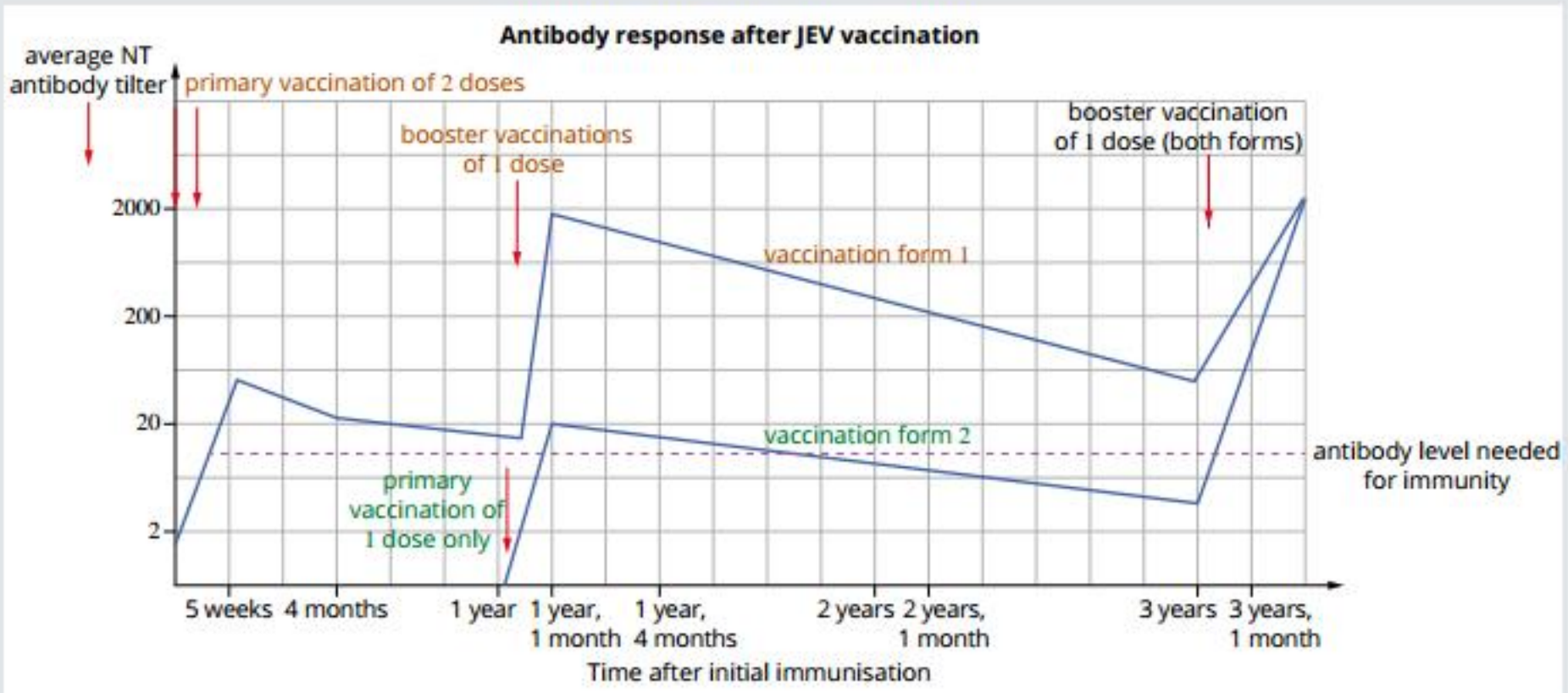
The antibody response of both options is shown in the graph. The dashed line represents the antibody level that is needed for protection from the virus.

Issues with the giving of the vaccination include cost (40 cents per dose) and the number of children who fail to complete the full series. Studies in India, which has a similar demographic to the country in question, on the drop-out rates for multidose vaccines, namely diphtheria, tetanus and pertussis (DTP) and polio oral vaccine (POV), are shown in the following graph.



When the vaccination program begins, initially 54 000 000 children under the age of 15 years will need to be vaccinated in a catch-up program. Thereafter, it is estimated that 3 150 000 children will need vaccination each year.

Prepare a recommendation for the east Asian government that outlines a vaccination program for their population. Provide a rationale for the JEV vaccination program.



Data analysis

DATA SET 1

The information below applies to Questions 1–5.
Tetanus is a serious disease for which vaccinations are available. Most people are vaccinated in childhood and many people do not have routine vaccinations for it later in life. A study was undertaken to determine how well protection was maintained after vaccination. In order to have long term protection, a patient should have an antibody titre of 1.0 TIU/mL. The results of the study are shown in Table 1.

TABLE 1 Antibody titres in the blood of subjects up to 15 years post-vaccination

Number of people tested	Number of tetanus vaccinations throughout their lifetime	Time since last vaccination	Median antibody titre (IU/mL)
17	Less than 5	≤5 years	1.85
12		6 ≤ 10 years	1.00
116		11 ≤ 15 years	0.51
206		>15 years	0.31
209	5	≤5 years	3.78
267		6 ≤ 10 years	2.21
102		11 ≤ 15 years	0.65
138		>15 years	0.47
189	More than 5	≤5 years	4.28
13		6 ≤ 10 years	2.23
23		11 ≤ 15 years	1.08
11		>15 years	0.67

Question 1 (1 mark)
What is the median antibody titre for the group containing a person who had their last vaccination 7 years previously and have had 5 vaccinations during their life?

Question 2 (2 marks)
Consider the data. Using the data what should the recommended maximum time be between doses? Justify your answer.

Question 3 (2 marks)
How does having more doses affect long term immunity? Use the data to answer.

Question 4 (2 marks)
In the group 6–10 years with 5 doses, one individual had an antibody level of 4.46 IU/mL. Explain this reading.

Question 5 (2 marks)
Why might the median be a better measure in this study than the mean antibody titres?

CHAPTER

09 Disease patterns

Epidemiology is the study of diseases, how they are transmitted, and how they might be controlled. Epidemiologists analyse disease patterns. Epidemiologists predict and control the spread of disease, by modelling and analysing transmission through communities, regions and across continents.

This chapter applies knowledge from previous chapters on disease and pathogens to a range of case studies. To develop models of disease transmission, this chapter introduces transmission concepts, mechanisms of transmission and methods of prevention.

The case studies provide in-depth and extensive learning activities that afford opportunities to use knowledge; incorporating retrieval and comprehension of knowledge, as well as analysis and interpretation of evidence. The data available in these chapters, enables students to apply and utilise knowledge and scientific skills beyond the questions and tasks. The case studies will develop skills to address all assessment objectives across all assessment instruments.

Syllabus subject matter

Topic 2 • Infectious disease and epidemiology

- Describe modes of disease transmission, including direct contact, contact with body fluids, contaminated food, contaminated water and disease-specific vectors. **9.1**
- Explain how the following factors affect the spread of disease
  - persistence of pathogens within host
  - transmission mechanism
  - proportion of the population that are immune or have been immunised
  - mobility of individuals in the affected population. **9.1**
- Interpret long-term immune response data. **9.2***
- Explain how personal hygiene measures, contact tracing and quarantine are used to control the spread of disease. **9.2**
- Analyse data to
  - predict outbreaks
  - determine the source of an outbreak
  - infer the mode of disease transmission
  - determine the effectiveness of different strategies in controlling the spread of disease. **9.2**

*The greyed-out dot point is also addressed in Chapter 8.

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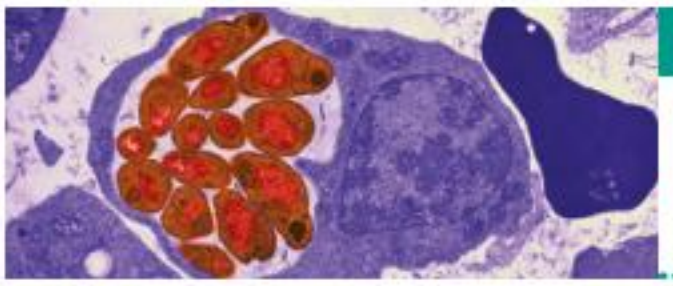
9.1 Transmission and populations



BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- describe the modes of disease transmission
- explain what factors affect the spread of disease and the speed of transmission.

The number of pathogens that exist in the world around you, including those on your body, is immeasurable. Fortunately, the microbiome on your body is relatively stable and pathogens usually need to gain access to the internal environment to cause disease.

TRANSMISSION AND SPREAD OF DISEASE

Disease itself cannot be transferred from one organism to the next; rather it is the pathogen that causes the disease that is transferred. You will recall from Module 7.2 the various ways in which pathogens can be transferred from one person to another, resulting in the transmission of disease. There are many mechanisms of transmitting pathogens, and the spread of disease becomes more complex the more people and organisms that are involved.

The spread of disease within a community is related to many factors, including population density and movement, personal and communal hygiene, immunity and environmental factors, mechanism of transmission and category of pathogen being spread. Epidemiologists sometimes study the spread of disease categorically to reduce the complexity. For example, when studying environmental factors, categories may include:

- water supply
- **sanitation** facilities
- food
- climate.

Each one of these environmental factors can be further analysed into more specific categories and then defined more discretely into measurable variables. Water supply factors can be analysed more specifically as inadequate excretion from disposal facilities, poor hygiene practices (such as hand washing, Figure 9.1.1), removal of used water from living premises, amount of stagnant water, or delivery of fresh water. Climate factors can be analysed further as flooding, heavy rains, excessive stagnated water and detritus, airstreams and wind.

FIGURE 9.1.1 Ultraviolet (UV) light being used to show bacteria on a person's hands. The person has applied gel to their hands and then washed them. Under UV light, the gel fluoresces (green) to indicate areas that have not been adequately cleaned. The UV light shows most species of bacteria regardless of whether they are harmful (pathogenic) or harmless.

The ability of pathogens to spread from one host to another and then through a community and global populations is continually developing. Even though much is known about pathogens, including their structures, chemistry, genetics, **virulence** and pathogenicity, we are still discovering more about them every year. Their ability to survive, the environments they can withstand, molecular interactions with hosts and environment are a few examples. Living pathogens require a medium to survive while being transferred and this usually includes water and nutrients. Non-living pathogens do not require water or nutrients. However, because they are not capable of movement, they are often transferred through mediums such as water, food, body fluids and direct contact of surfaces. As long as the non-living pathogens remain structurally sound, and are not denatured by heat or extreme conditions, they will remain pathogenic.

Living organisms are the perfect vectors to transport and spread pathogens and transmit disease: they have a reasonably stable temperature, contain water and fluid, are a source of nutrients, and typically move regularly. Of all the vectors that spread disease to new populations, humans are the most potent. To develop an understanding of human influence on disease transmission, research into international movement showed that in the 1990s, 500 million people travelled across international borders on commercial flights. The same research showed that over a 200-year period, the average daily distance travelled by the typical French population increased 1000-fold, and that in 40 years the number of people who exited or entered Australia increased 100-fold. Figure 9.1.2 shows the traffic of the world's population in recent times.

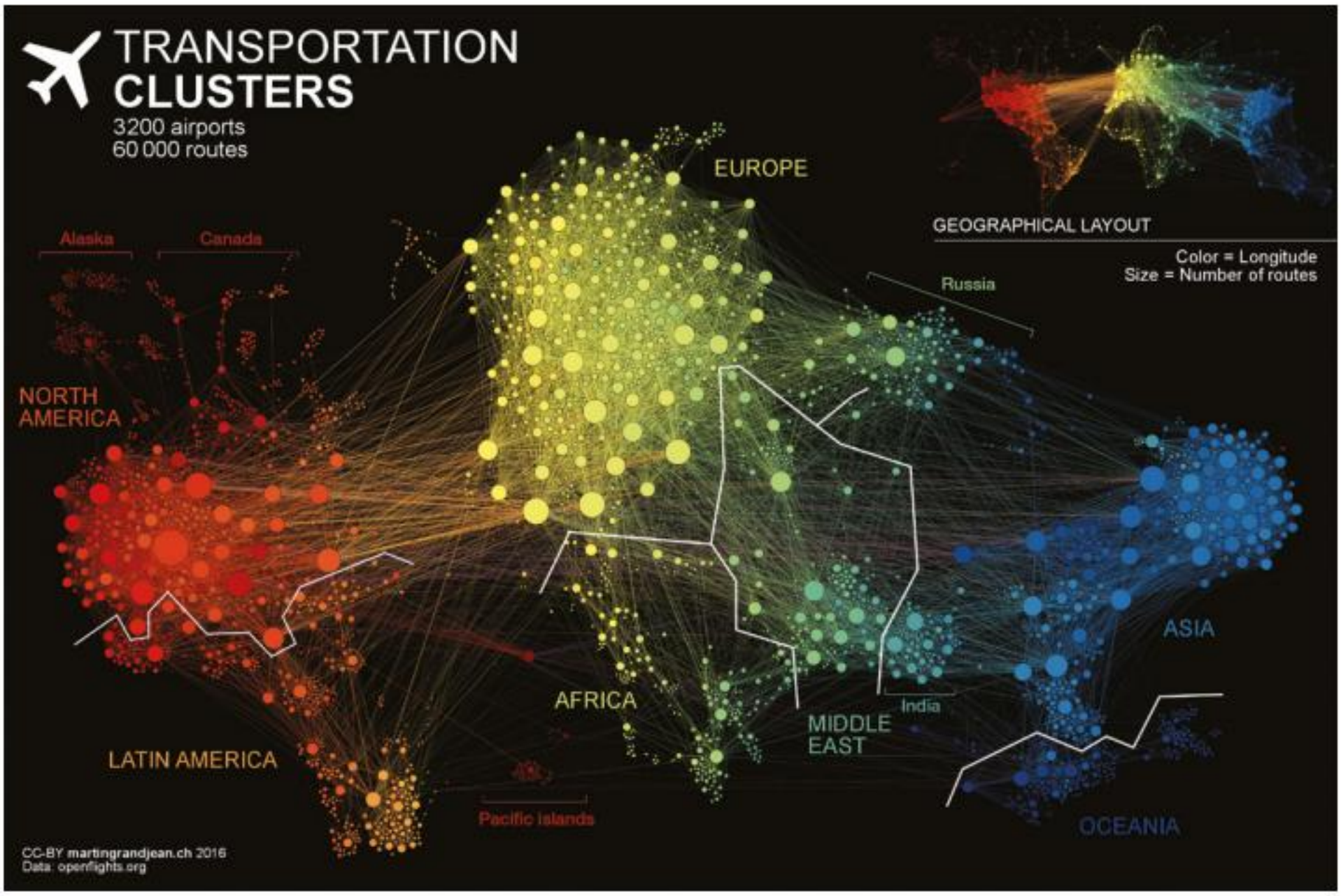


FIGURE 9.1.2 This map displays the 60 000 flight routes of 2016 (shown by size of the circle at each hub), representing the global reach of air travel (or longitude—indicated by the colour). It is estimated that the number of daily flights was in excess of 100 000 in 2016, equating to more than 8 million people travelling by air.

Table 9.1.1 outlines a few of the pathways in which disease has been shown to spread because of human activity.

TABLE 9.1.1 A few of the recorded pathways in which diseases have been known to spread

Pathway	Vector	Direct/ indirect	Transmission mechanism
fishing routes	human	both	• person-to-person (skin–skin) • contaminated object (surface–orifice) • animal-to-person (fluid transfer) • person-to-person (fluid transfer)
trading routes	human	both	• person-to-person (skin–skin) • contaminated object (surface–orifice) • animal-to-person (fluid transfer) • person-to-person (fluid transfer)
	human and snail	indirect	• vector (human–snail–human)
migration	human	both	• person-to-person (skin–skin) • contaminated object (surface–orifice) • person-to-person (fluid transfer) • fluid transfer (through food, water supply, ventilation)
	insect (sandfly)	indirect	• fluid transfer (through food)
shipping	human	both	• skin–skin • surface–orifice • fluid transfer (human–human)
	insect (mosquito)	indirect	• fluid transfer
	various introduced organisms	indirect	• environmental reservoir • food and drinking water
infrastructure	human	indirect	• airborne (ventilation systems) • food and drinking water

Table 9.1.1 is not an exhaustive list of recorded pathways for disease transmission. Importantly, of all vectors and disease transmission pathways, humans are the most common.

Mechanism of transmission

To survive, all living pathogens need a source of nutrients and many require a specific source. Many living pathogens that cause disease must infect new people, animals or plants. As disease causes the malfunctioning of processes in a living host, if the disease persists in the host, then often it is only a matter of time before the host dies. For living pathogens, the mechanism of transmission must sustain life until a new host is found. Some pathogens are highly resistant to environmental factors and can remain alive under harsher conditions for longer than other pathogens. For example, the spores of *Bordetella pertussis*, the bacterium that causes whooping cough, can survive for many days in the environment. Non-living pathogens are usually limited by fewer environmental factors in transmission.

The mechanisms of transmission are grouped into two distinct categories: direct and indirect contact. **Direct contact transmission** is when disease is transferred from an infected person to a **susceptible** person through physical contact. Direct contact includes person-to-person (direct touch or exchange of body fluids) and droplets (fluid droplets sprayed by coughing, sneezing or speaking). Figure 9.1.3 shows some examples of methods of direct contact transmission. **Indirect contact transmission** is when disease is transferred to a susceptible person where no physical contact has been made. Indirect contact includes airborne transmission, contaminated objects, food and drinking water, animal-to-person contact, vectors and environmental **reservoirs**.

Person-to-person transmission

The skin or affected body parts have pathogens present that are transmitted directly through hands, clothes, hair or any other contaminated material through physical contact. If the pathogen requires an internal or wet environment to live, it will enter the body through broken skin, **mucous membranes** (e.g. lips) or an orifice. Often these infections are associated with poor personal hygiene.

Because transmission must be through direct physical contact, the pathogens do not travel very far, as their motility is limited. Transmission is achieved directly through the actions of the host. Transmission through person-to-person contact can be traced through very small transmission distances and usually to people of regular contact, such as family members, carers or perhaps work colleagues. **Disease clusters** appear in or around the home of the infected, and the homes of their family members or friends, carers or work colleagues (Figure 9.1.3).

FIGURE 9.1.3 (a) Skin-to-skin contact or breathing the same droplets from exhaled air cause direct contact transmission. (b) Skin-to-skin contact, droplets, direct contact of clothing or sharing eating and drinking utensils can result in direct contact transmission. (c) Large numbers of sick people in a small area can result in direct contact transmission through clothing and droplets (breathing and coughing). (d) Sharing of facilities at the same time will result in skin-to-skin contact and direct contact transmission through droplets (breathing and perhaps coughing or sneezing). (e) Couples easily transmit disease through direct contact of clothes, skin-to-skin contact and body fluid transfer when kissing or through sexual relationships.

FIGURE 9.1.4 Sneezing ejects and transfers droplets 1–2 metres, demonstrating direct contact transmission of disease.

Droplet transmission

When an individual talks, coughs or sneezes, small **droplets** of mucus, which contains pathogens, are expelled or ejected (Figure 9.1.4). When droplets reach a susceptible individual, the person may inadvertently transfer the pathogen to a skin opening, mucous membrane or an orifice (e.g. using their hands to scratch an itch).

Droplet transmission depends on the distance the droplets travel when ejected. The distance the droplet travels depends on the size of the droplet and the ejection velocity. A transfer distance of 1–2 metres is typical for droplets. Droplet transmission under 2 metres is considered direct contact transmission.

Disease clusters for droplet transmission are similar to person-to-person contact, although it is possible for more friends or work colleagues to become infected. Usually when an infected individual becomes aware of their illness, they remain at home, minimising the chance of transfer. This can result in a cluster similar to person-to-person contact because only family, close friends or carers spend time with infected individuals once they are aware.

Airborne transmission

If a pathogen can remain in the air for a long period of time and can travel more than 1–2 metres, it is classified as airborne. Airborne pathogens can travel further than pathogens carried by droplets, usually transferring diseases within closed facilities or buildings to other rooms or areas of the building. They can also remain in the air for a long time, so pathogens can be transferred to susceptible individuals after an infected person is gone. Often pathogens are carried on dust or fine particles, known as aerosols, and can travel large distances over long periods of time.

Disease clusters for airborne pathogens can often be found at locations where infected individuals have visited. Often a person will visit a site while infectious but before symptoms are present. This means transfer can occur at many locations and affect many people following an infected person's movements. The clusters can be traced along movement patterns and may develop a timeline. Measles (rubeola) virus is an airborne pathogen that is found in tiny droplets that remain in the air for extended periods of time and can travel large distances of up to 10 metres (without assistance from wind or airflow). Figure 9.1.5 illustrates the transfer of the measles virus.

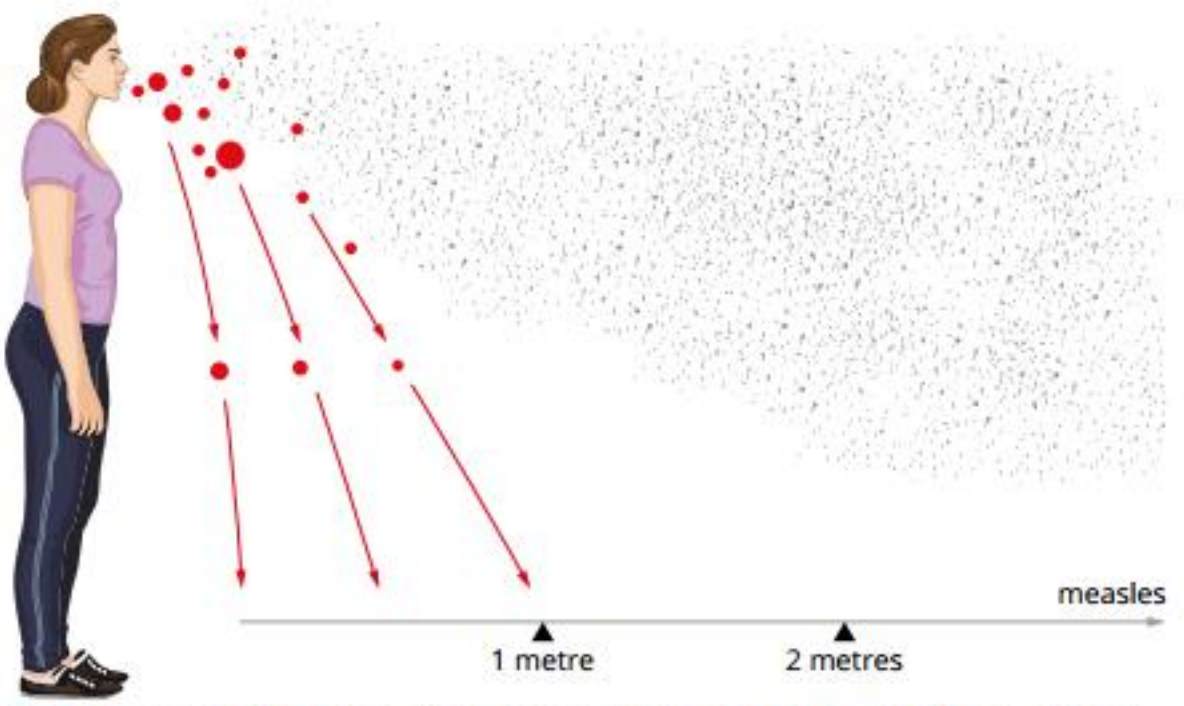


FIGURE 9.1.5 Measles (rubeola) virus is an airborne pathogen that is found in tiny droplets and remains in the air for extended periods of time and can travel large distances.

Transmission through contaminated objects

Infected individuals can eject droplets or touch inanimate objects, transferring the pathogen to the object. Some of these objects allow the pathogen to live or can transfer infectious pathogens to susceptible individuals. Objects or materials that can carry pathogens are called **fomites**. Fomites do not sustain living pathogens indefinitely and the window of opportunity for infection is limited. When a susceptible individual touches the fomite and receives the pathogen, they then usually need to transfer the pathogen to a point of entry for transmission to occur. A computer keyboard is an example of a fomite (Figure 9.1.6), transferring bacteria from an infected person to another. The recipient may then transfer the bacteria to an opening by rubbing their eyes or nose.

Although disease transmission and spread through fomites can be traced, it is difficult to assert with confidence. Health and disease organisations and professionals can ascertain the presence of pathogens at a location, but many variables affect whether it was transmitted. Such variables include personal and organisational hygiene practices, climate, airflow facilities and possible effective contact.

FIGURE 9.1.6 Ultraviolet light showing bacteria (illuminated in green) on a computer keyboard

Transmission through food and drinking water

Food and water (especially drinking water) are known as vehicles to transport pathogens (Figure 9.1.7). The sharing of food and water with an infected person, including before they realise that they are infected, can transmit disease directly to the internal environment of a susceptible person. Poor sanitation methods (both personal hygiene and community infrastructure) allow pathogens to be transported great distances.

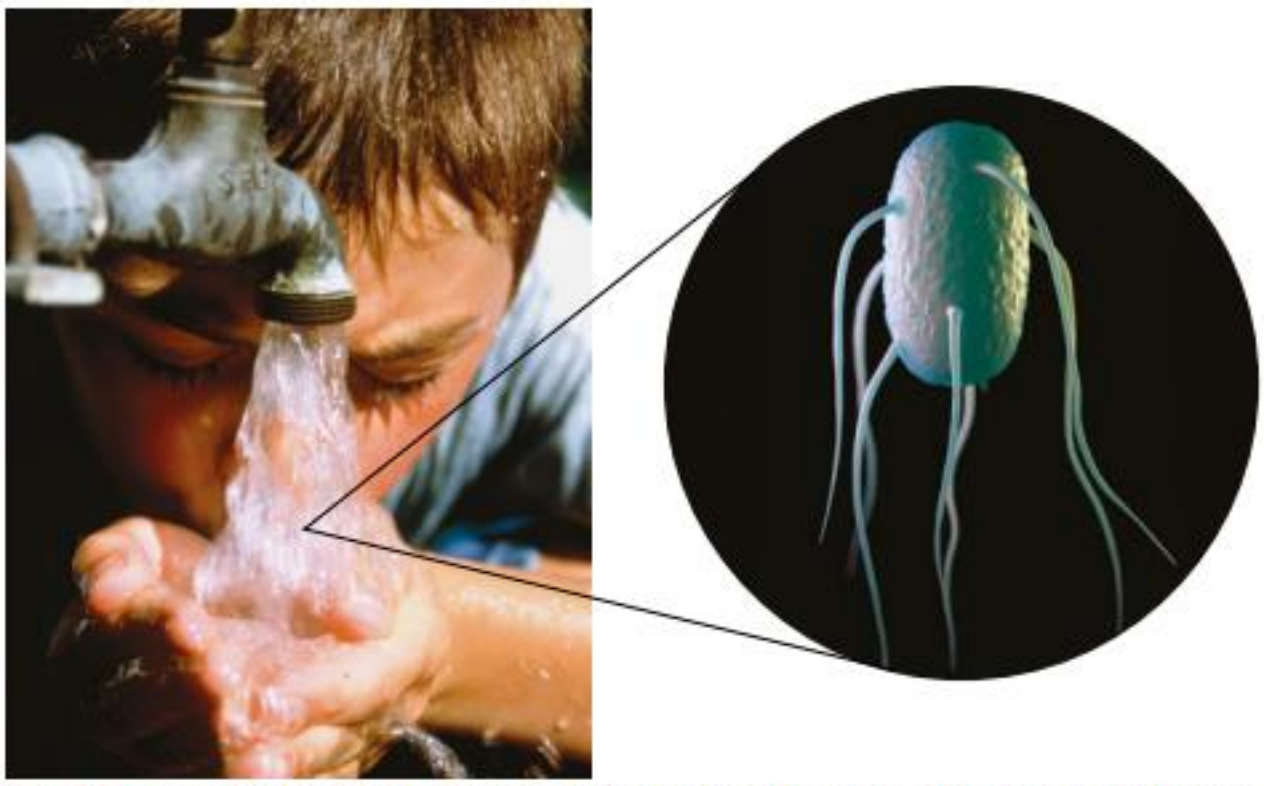


FIGURE 9.1.7 This boy is drinking water contaminated with *Salmonella* bacteria. Many countries do not have clean or filtered drinking water; disease from unboiled water is common.

Animal-to-person transmission

Animal-to-person contact transmission of infection is similar to person-to-person direct contact. Some pathogens can infect and cause disease in both animals and humans. The transfer may occur through a scratch or bite as well as from skin to skin. Figure 9.1.8 shows the results of two animal bites.

FIGURE 9.1.8 (a) This animal bite has resulted in a slight infection. (b) This bite has caused a more severe infection.

Vectors

FIGURE 9.1.9 Mosquitoes can carry numerous pathogens because they feed on the blood of hosts, transferring blood containing pathogens from one person to another.

Vectors are organisms that transfer a pathogen from an infected individual to a susceptible individual. Common vectors are insects such as mosquitoes (Figure 9.1.9), ticks, fleas and lice, also rats, cattle, cats and dogs. Often vectors carry the pathogen but are not diseased. Insects often draw fluid (e.g. blood) from an infected host and then move on to another person who may be susceptible, thus transferring the pathogen.

Researchers are interested in the route of vector transmission, some of which can be quite complicated. For example, pathogens from infected individuals can be found in faecal matter or waste, which vectors may access or feed on. Pathogens are then transferred when the vector comes into contact with a susceptible host. Other routes involve vectors increasing in number during certain seasons, or when particular environmental conditions exist. Some routes include vector-to-vector transfer before reaching a human host. As human population density increases, so does the number of possible links in the transmission chain, enabling pathogens, and the disease, to spread further.

To consider disease transmission via vectors, the life cycle of the vector also needs to be studied. As such, treating disease spread by vectors can be significantly influenced by treating the vector itself. For example, the *Plasmodium* parasites that cause malaria are spread by mosquitoes; the use of insect repellent and guarding against mosquitoes in malaria-prone environments helps to reduce the spread of malaria. The Lyme disease cycle in Figure 9.1.10 illustrates how the bacterium *Borrelia burgdorferi* is transferred through the tick vector. The cycle is complex and involves many organisms and opportunities to transfer to humans. This cycle is 2 years long and is seasonal, with the seasons colour-coded in the diagram: spring (green), summer (yellow), autumn (orange) and winter (blue-grey). In some instances, the complexity of the transmission route can also include a vector transferring the pathogen to fomites or other animals, which eventually come in contact with a susceptible human.

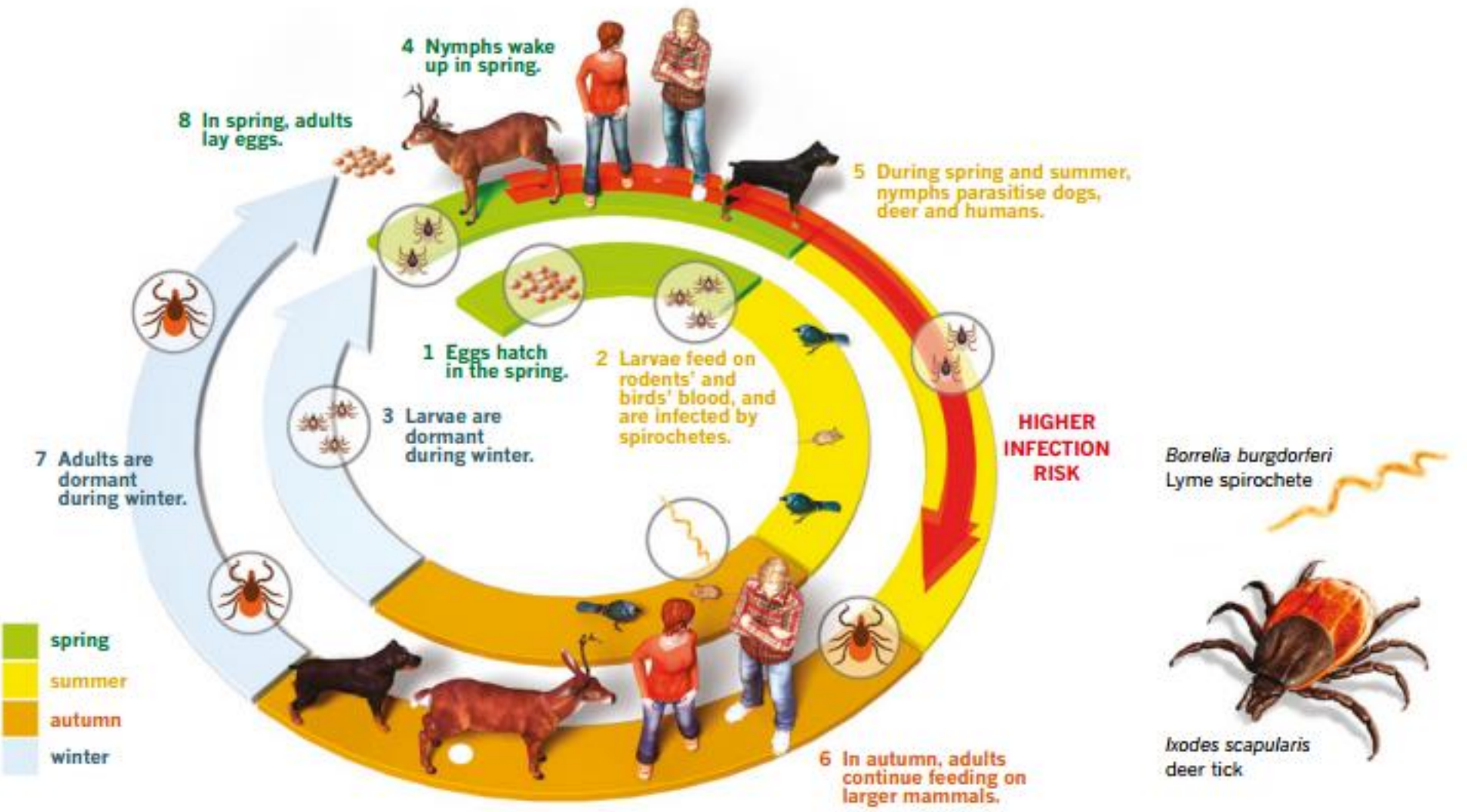

4 Nymphs wake up in spring.
8 In spring, adults lay eggs.
5 During spring and summer, nymphs parasitise dogs, deer and humans.
1 Eggs hatch in the spring.
2 Larvae feed on rodents' and birds' blood, and are infected by spirochetes.
3 Larvae are dormant during winter.
7 Adults are dormant during winter.
HIGHER INFECTION RISK
6 In autumn, adults continue feeding on larger mammals.
spring
summer
autumn
winter
Borrelia burgdorferi
Lyme spirochete
Ixodes scapularis
deer tick

FIGURE 9.1.10 Diseases that are transferred through vectors can have complex cycles and pathways. This Lyme disease cycle illustrates how the bacterium *Borrelia burgdorferi* (right) is transferred through the tick vector.

Disease clusters for vector-spread pathogens can be found around communities with poor sanitation, and near vector population hot spots and breeding grounds. The most prolific vectors are humans. The increasing regional and global movement of humans is causing new patterns of disease clusters and transmission routes.

Environmental reservoirs

Reservoirs in the environment can be anything that can harbour or store pathogens or vectors, such as the water reservoirs in Figure 9.1.11. Soil, water and vegetation can all harbour pathogens or vectors, which can be transmitted to people either directly or indirectly. As this is location based, clusters of disease typically occur in relation to a location.

Regional spread of disease

The spread of disease in local regions has always been considered closely linked to distance. An example of the relationship between distance and disease transmission is the Philadelphian typhoid epidemic in the USA in the late 1800s to early 1900s. Clusters of typhoid formed along water supply infrastructure and in households, maintaining a correlation between the source and distance. In recent times, the spread of disease has increasingly correlated to transport systems, including road networks and airports. Disease is also known to spread through social networks, and spread is affected by accessibility to source, infrastructure contamination and transport of the pathogen. Factors that can affect the rate of infection and transmissibility include temperature and humidity, precipitation, airflow and ventilation, hygiene practices and human behaviour.

Disease vehicles can cause both localised and regional clusters of infected individuals. The clusters may follow infrastructure systems (as occurred during the Philadelphian typhoid epidemic discussed above). Clusters can also be seen along travel routes (air and sea where food is served or shared) or in places of large gatherings (e.g. cruise ships and conferences).

The spread of disease is due to many factors and research into a local case at the location of an outbreak reveals numerous methods and mechanisms of transmission that are often unique to the case. Although the exact details of all transmissions in a case cannot be determined, models are developed based on recorded data to infer a likely scenario and plausible explanation. Many similarities are found between cases, though it is rare to find two identical cases of disease transmission. Hence, distance is often (though not always) a factor that is most often strongly correlated to disease transmission. Data must be analysed, along with maps and transport systems to try to interpret and understand the methods and spread of disease.

Global spread of disease

As modes of transport, including air, sea and land, continue to develop, the speed of travel increases as well as the number of people travelling. This means all vectors, especially humans, and pathogens are moving further, faster and in greater numbers. Notable increases in transport are shown in Table 9.1.2. Advances in transport have also caused a noticeable increase in the global contact of flora and fauna between continents and regions, especially by individuals from wealthy nations. Growth across the globe in human migration, tourism and economic activity is leading to continual increases in the movement of both disease vectors and the number of diseases.

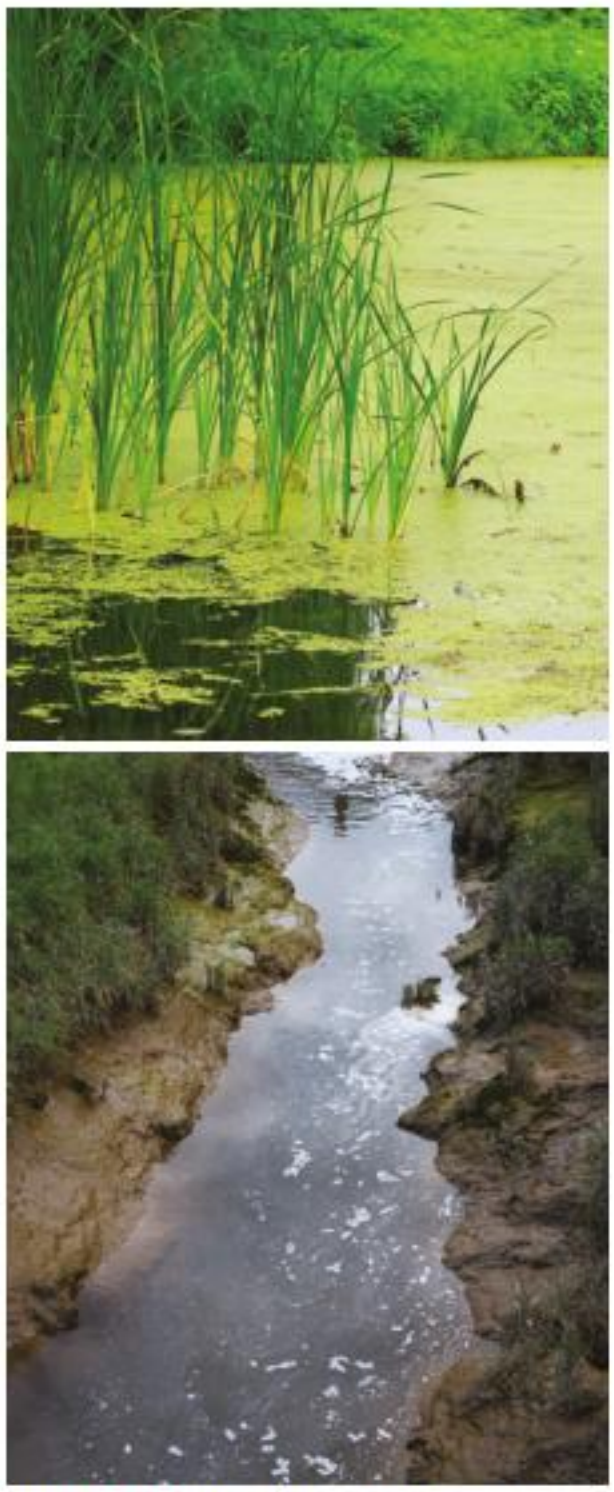


FIGURE 9.1.11 Water reservoirs provide both nutrients and a reproductive environment for pathogens and vectors.

TABLE 9.1.2 Increases in worldwide transport and associated vectors

Transport industry	Increase	Time	Transported vectors and/or disease
commercial airline passengers	9% per year	since 1960	• severe acute respiratory syndrome (SARS) • cholera • influenza • yellow fever • dengue • malaria
airfreight	about 9% per year	since 1960	• rats • mosquitoes (malaria) • animals such as pets
shipping	27%	since 1993	• fleas • lice • kissing bugs • mosquitoes (especially *Aedes aegypti, Culex pipiens complex, Aedes albopictus)*

MEASURING THE SPREAD OF DISEASE

Organisations concerned with local and worldwide disease and health include the World Health Organization (WHO) and the Centres for Disease Control and Prevention (CDC). WHO and the CDC have published many articles correlating global economic activities, as well as tourism and human migration, to an increasing number of cases in the movement of both disease vectors and the diseases they carry worldwide. Historically, trade routes, seaports, exploration and travel by horseback or on foot through villages have been strongly correlated to the spread of disease and **pandemics** (outbreaks of diseases across large regions). Animal vectors can also contribute to or sometimes cause pandemics. The Australasian Epidemiological Association (AEA) is responsible for monitoring health practice, teaching and research within Australasia.

Rate of disease transmission: R_0

Because of the complexity of disease spread, transmission is usually measured first by the incidence of infections and rate of disease spread, not the mode of transmission or other factors. The ability of a disease to spread, and how infectious it is, is measured by the basic **reproduction number (R_0)** (pronounced 'R nought'). R_0 is a measure (or estimation) of how many people will contract a disease from the person who is the primary source. R_0 is the number of **secondary infections** produced by a single case of infection, in a population where the disease does not exist and therefore where individuals in the population are vulnerable. This is shown in Figure 9.1.12.

R_0 is measured by counting the number of secondary cases following the introduction of an infection into a totally susceptible population. Three parameters are considered when calculating this number: The duration of contagiousness, the likelihood of infected people and susceptible people, and the contact rate. If R_0 is 7, it means that approximately seven people will become infected from coming into contact with a diseased person. Subsequently, every new case of the disease will result in seven new secondary cases. The R_0 of several infectious diseases can be seen in Table 9.1.3. R_0 can be used to predict outbreaks and pandemics. When:

- $R_0 > 1$, then transmission is increasing, and an outbreak is possible
- $R_0 \approx 1$, then transmission is stable
- $R_0 < 1$, then transmission is decreasing, and the disease is going into extinction (in the local region).

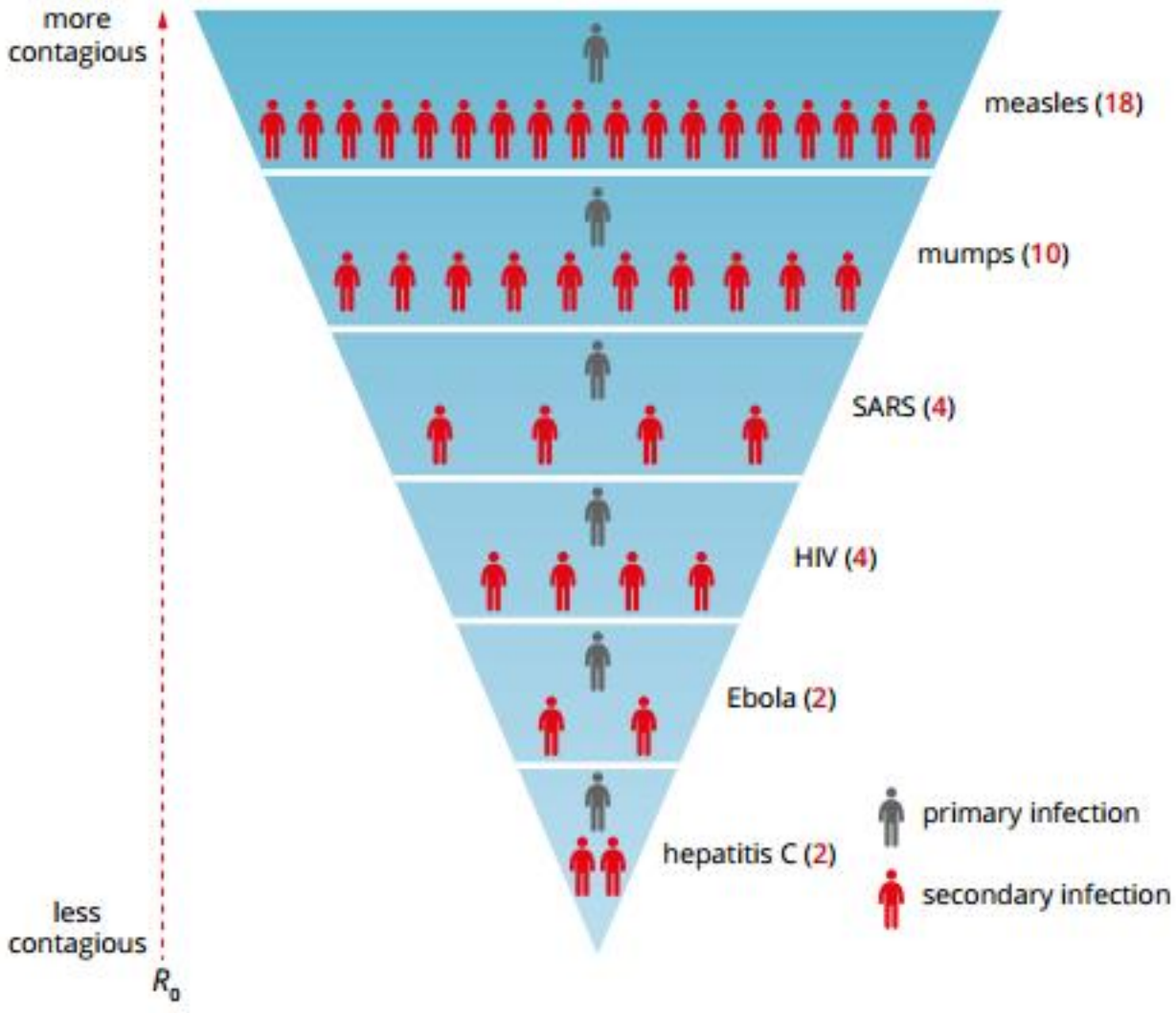


FIGURE 9.1.12 R_0 for some diseases. The numbers of secondary infections that are typically caused by a single person are shown in parentheses.

TABLE 9.1.3 Estimated R_0 values for some well-known or common infectious diseases

Disease	Mechanism of transmission	R_0
measles	airborne	12–18
diphtheria	droplet	6–7
small pox	airborne	5–7
rubella	airborne	5–7
mumps	airborne	4–10
HIV/AIDS	person-to-person	2–5
pertussis (whooping cough)	airborne	5.5
SARS	airborne	2–5
influenza (1918 pandemic strain)	airborne	2–3
Ebola (2014 Ebola outbreak)	person-to-person	1.5–2.5

Disease does not always spread when people come into contact with or are in the proximity of transmission. Only some people may fall ill, resulting in a secondary infection, as shown by the R_0 values.

Several factors affect the rate of secondary infections, and the calculation of R_0 depends on the:

- length of time an individual is infectious
- probability of transmitting the infection to a susceptible individual during a single contact
- rate of new individuals contacted.

The estimated R_0 is not always the same for a specific disease across locations or over time. This is because the factors that affect the calculation of R_0 change. For example, a disease may have a larger R_0 value in a densely populated area than in a sparsely populated area. Also, different strains of the same disease may have different levels of virulence, which will affect the calculated R_0. Standards of hygiene, education and infrastructure can also affect calculated R_0 values. Likewise, not all individuals who fall ill require medication or seek medical aid, so some R_0 values are measured from recorded cases while others are estimated. Table 9.1.4 shows the R_0 values for two strains of influenza at different times and locations.

TABLE 9.1.4 Estimated R_0 for the H5N1 strain of the influenza virus

Year(s)	Subtype	Location	R_0
1976	H1N1	New Jersey, USA	1.20
2004–2006	H5N1	Vietnam	0.00
2004–2006	H5N1	Indonesia	0.00
2005	H5N1	Turkey	<1
2009–2009	H5N1	Indonesia	0.1–0.25
2006	H5N1	Indonesia	1.14

Pathogen persistence

One of the many factors that influence the spread of disease is the persistence of pathogens to infect a host. Within a host, pathogens can lie dormant for extended periods of time and remain inactive until triggered, or they can have various periods of time in which they are infectious. Each pathogen has its own infectious characteristics due to reproduction rate, toxicity, virulence and other factors. Tables 9.1.5 and 9.1.6 show the length of time some bacterial and viral pathogens persist.

When analysing a case of disease transmission, it is very important to identify the pathogen. It is important to know the pathogen and understand its characteristics and mechanisms of transmission. Knowing such factors helps researchers studying patterns in the spread of disease.

REDUCING TRANSMISSION

You will recall that in Chapter 8 specific details about innate and adaptive immunity as well as vaccinations and immunisation were explained. Immunity and immunisation programs have been shown to decrease disease transmission and decrease the spread of disease through communities as well as across regional and international borders. Many methods and strategies are used to decrease the transmission and spread of disease, including medicine and personal health care, disinfectants and aseptic techniques, personal hygiene and infrastructure sanitation, food and water supply, using pesticides and killing vectors. However, the most effective method and strategy to control the transmission and spread of disease has been immunisation.

TABLE 9.1.5 The length of time various bacteria can remain pathogenic on inanimate objects

Bacterium	Disease(s) caused by bacterium	Duration of persistence
Acinetobacter spp.	pneumonia, meningitis	3 days to 5 months
Bordetella pertussis	whooping cough	3–5 days
Campylobacter jejuni	campylobacteriosis	up to 6 days
Clostridium difficile (spores)	colon infection	5 months
Chlamydia pneumoniae, C. trachomatis	chlamydia	≤30 hours
Chlamydia psittaci	chlamydia	15 days
Corynebacterium diphtheria	diphtheria	1 week to 6 months
Escherichia coli	gastroenteritis	1.5 hours to 16 months
Helicobacter pylori	peptic ulcers, gastritis	≤90 minutes
Klebsiella spp.	pneumonia	2 hours to >30 months
Listeria spp.	listeriosis (food poisoning)	1 day to months
Mycobacterium bovis	tuberculosis (TB)	>2 months
Mycobacterium tuberculosis	tuberculosis (TB)	1 day to 4 months
Neisseria gonorrhoeae	gonorrhea	1–3 days
Proteus vulgaris	urinary tract infections	1–2 days
Salmonella typhi	typhoid fever, food poisoning, gastroenteritis	6 hours to 4 weeks
Staphylococcus aureus, including methicillin-resistant S. aureus (MRSA)	impetigo, food poisoning, cellulitis, toxic shock syndrome	1 week to 7 months
Streptococcus pneumonia	meningitis, peritonitis, sinusitis	1–20 days
Vibrio cholera	cholera	1–7 days

TABLE 9.1.6 The length of time various viruses can remain pathogenic on inanimate objects

Virus	Duration of persistence
adenovirus	7–90 days
astrovirus	7–90 days
coronavirus	3 hours
SARS associated virus	3–4 days
coxsackie virus	>2 weeks
cytomegalovirus	8 hours
echovirus	1 week
hepatitis A virus (HAV)	2 hours to 60 days
hepatitis B virus (NBV)	>1 week
human immunodeficiency virus (HIV)	>1 week
Herpes simplex virus, type 1 and 2	4.5 hours to 8 weeks
influenza virus	1–2 days
norovirus and feline calici virus (FCV)	8 hours to 1 week
papillomavirus 16	>1 week
papovavirus	8 days
parvovirus	>1 year
poliovirus type 1	4 hours to 8 days
poliovirus type 2	1 day to 8 weeks
pseudorabies virus	≥7 days
respiratory syncytial virus	up to 6 hours
rhinovirus	2 hours to 7 days
rotavirus	6–60 days
vacciniavirus	3–20 weeks

Population/herd immunity

You will recall that Chapter 8 explained how innate and adaptive immunity develops to form antibodies that provide a highly tuned response to infection by pathogens. Individual immunity can minimise or prevent disease even after acquiring a pathogen. Immunity that is achieved by a critical portion of a community (i.e. a critical number of the population) that results in most individuals in the community being protected against a disease is called **community immunity**, or herd immunity. The critical number of the population required to achieve herd immunity depends on many factors, such as the R_0, population susceptibility and mechanism of transmission.

Herd immunity is achieved by exposing a critical portion of the community to a pathogen, or modified strain of a pathogen, so each individual develops a personal immunity. Personal immunity decreases the time a pathogen is infectious within a host and also decreases the number of pathogens that reproduce within the host. Together, these factors decrease the ability to infect and spread a disease, helping to maintain the health of the wider community.

Because many factors influence disease transmission and humans are the most potent vectors, human preventative measures are the most successful way of combating disease transmission. Research shows the most effective, efficient and safe way to achieve this is through vaccines. The more people who are vaccinated, the less chance there is of an infectious pathogen spreading throughout a population. Herd immunity results in less disease in the community, less severe infections (in general) and protection for individuals for whom other methods of protection are too risky, including the young, elderly, pregnant, ill and **immunosuppressed**. Achieving immunity of a critical portion of a population changes the status of the population from susceptible to protected.

Figure 9.1.13 shows how herd immunity works. An infected person will come into contact with people through their daily schedule. Depending on the R_0, a number of secondary infections will occur. R_0 decreases significantly when large portions of a community are immunised, thus protecting most of the community.

The people in a population who are at higher risk of disease and benefit from herd immunity are:

- young children—their immune systems (innate and adaptive) are still developing as they grow, and disease often strikes this portion of the population with greater severity and higher **morbidity** rates
- the elderly—their immune systems become less efficient and effective as people get older and so elderly people are more susceptible to disease
- pregnant women—their immune system is suppressed during pregnancy; a large immune system response during pregnancy can cause deformities or fatally harm the growing foetus
- individuals who are already ill—these people have a suppressed immune system (which can also be caused by medication), so are more susceptible to disease.

For these reasons, herd immunity can protect most people in a community.

In 1980, WHO declared that one of the world's deadliest diseases, smallpox, had been eradicated as a result of vaccines and herd immunity. Smallpox (variola virus) is transmitted by droplets (direct contact transmission). The last known natural case was in Somalia in 1977.

Immunisation coverage

Immunisation coverage is the proportion of the population that has been vaccinated against a particular disease. Epidemiological studies and organisations attempt to calculate the immunisation coverage required to protect the rest of the community as illustrated in the example in Figure 9.1.13.

Figure 9.1.13 displays **herd immunity thresholds** required to protect a community from disease outbreaks in common diseases. The R_0 values are provided as a range due to the multiple variables involved in disease transmission. Even though many variables are involved, the R_0 value is somewhat consistent across numerous communities and continents. Also notable is the correlation between R_0 and herd immunisation threshold.

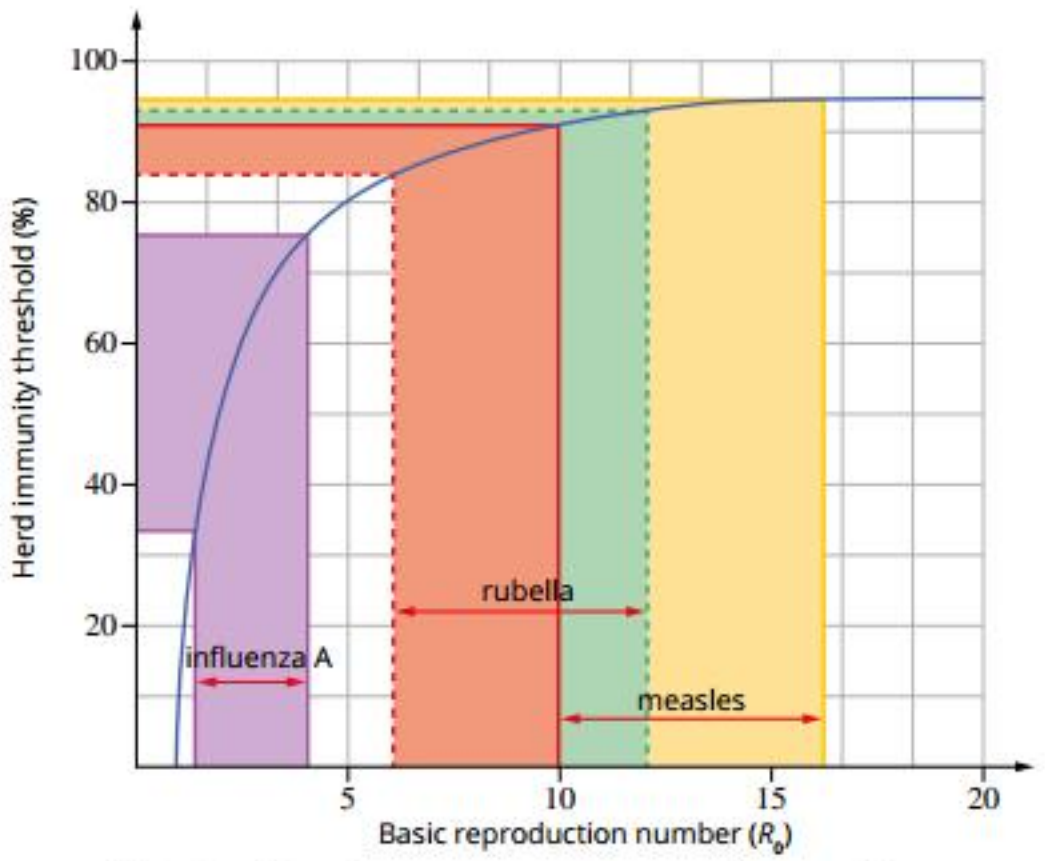


FIGURE 9.1.13 Using typical R_0 values across numerous community cases of disease spread for influenza A, rubella and measles, immunisation coverage has been estimated (modelled) in order to determine a proposed critical portion of the community (herd immunity threshold) that would protect the rest.

Table 9.1.7 illustrates the correlation between vaccination (immunisation) coverage and R_0 values for common diseases.

Table 9.1.8 shows the result of a study of the spread of mumps across numerous primary schools in the Netherlands. There is a general trend in the overall attack rate (number of people diagnosed with the disease per population sample) that correlates with the vaccination coverage. As can be seen from the data, the correlation between overall attack rate and vaccination coverage is similar to the estimates of R_0 in Table 9.1.7. The data implies that herd immunity can be achieved at about 78% vaccination coverage or higher.

TABLE 9.1.7 Calculated estimates for the immunisation coverage needed in a population to protect the community from outbreaks

Disease	R_0	Herd immunity threshold (%)
diphtheria	6–7	85
measles	15–16	83–94
mumps	4–7	75–86
pertussis	12–17	92–94
polio	5–7	80–86
rubella	6–7	83–85
smallpox	5–7	80–85

TABLE 9.1.8 The spread of disease (mumps) across ten primary schools in the Netherlands

	Number of people	Number infected	Vaccination coverage	Attack rate in unvaccinated people	Attack rate in vaccinated people	Overall attack rate
All schools	2493	510–1342	0.62	0.68 (485/709)	0.03 (25/952)	0.31
School 1	432	205–369	0.12	0.86 (204/237)	0.03 (1/31)	0.76
School 2	338	135–289	0.13	0.82 (131/160)	0.17 (4/24)	0.73
School 3	259	68–159	0.42	0.72 (68/94)	0 (0/74)	0.40
School 4	184	40–70	0.54	0.53 (37/70)	0.04 (3/84)	0.26
School 5	130	13–33	0.75	0.46 (13/28)	0 (0/82)	0.12
School 6	263	28–171	0.76	0.70 (19/27)	0.10 (9/93)	0.23
School 7	194	6–43	0.78	0.19 (6/31)	0 (0/126)	0.04
School 8	227	3–27	0.79	0.05 (2/41)	0.01 (1/162)	0.01
School 9	258	6–119	0.93	0.18 (2/11)	0.03 (4/134)	0.04
School 10	208	6–62	0.93	0.30 (3/10)	0.02 (3/142)	0.04

CASE STUDY 9.1.1

Measles outbreak in Western Sydney, April 2017

In April 2017, the New South Wales Government Department of Health (NSW Health) reported a measles outbreak in Western Sydney. The number of confirmed cases of locally acquired measles cases associated with the outbreak was 16.

The cause of the locally acquired cases was most likely a single person who was infected with the disease in Indonesia and spent time in the Western Sydney region while infectious.

In the latest case, a man who was infected by an earlier case at Quakers Hill on 28–30 March, spent time in Lawson, Strathfield and Leichhardt on 10 April, and later on 14 April went to the Blue Mountains Hospital where he was isolated and recovered.

On the day of 10 April, he visited the Uniting Marion Aged Care Facility in Leichhardt, the Me Oi Vietnamese restaurant in Strathfield, and the Lawson shops in the Blue Mountains. He revisited the shops on 13 April. Typical infections result in a rash on day 14, and infected people are usually contagious from 4 days before until 4 days after the onset of the rash. The infected individual presented themselves to the hospital on 14 April, the third day he had a rash.

Vicky Sheppeard, Director of Communicable Diseases at NSW Health, released a press statement regarding this case, stating that isolation of infected individuals is the best method to combat any further outbreak.

Review

1. State the mechanism of transmission for measles. (Hint: Use Module 9.1.)
2. Outline the range of dates from Table 9.1.9 in which people could have been infected and contracted the measles in this case.
3. Predict the susceptible populations from Table 9.1.9 that could be at risk from this case. Support your prediction with adaptive immunity concepts.
4. NSW Health issued a statement releasing the details of all locations visited by the latest case (Table 9.1.9). Knowing that the infection was transmitted in Quakers Hill, explain why it was important to release this information.
5. Write an appraisal of Vicky Sheppeard's press release regarding isolation by detailing implications and limitations.

TABLE 9.1.9 The timeline of all locations visited by the latest infected case of measles in Western Sydney

Date	Location
18 April	Casula Central Medical Centre and Chemist Warehouse, Casula approx. 10am
15 April	Blacktown Hospital, approx. 7.30–8 pm
13, 15 and 17 April	Liverpool Westfield, including an optometry practice
14 April	Rashays, Darling Harbour (later in the afternoon)
14 April	Powerhouse Museum (early afternoon)
10 and 13 April	Lawson shops, Lawson
10 April	Uniting The Marion Aged Care Facility, Leichhardt
10 April	Me Oi Vietnamese restaurant, Strathfield
8 April	Virgin flight VA965, Sydney to Brisbane
7 April	Local train and bus travel in Western Sydney area
28 March to 4 April	Auburn area, including Pharmacy 4 Less
3 April	NAS Advanced Medical Centre, Auburn
1 April	Michel's Patisserie, Auburn
1 and 2 April	The Children's Hospital emergency department
1 April	Fairfield Hospital emergency department
28–30 March	Wyndham College, Quakers Hill
28–30 March	Local train between Flemington and Quakers Hill
26 and 31 March	Fairfield District Medical Centre
26 and 28 March	Tweed Hospital emergency department, Tweed Heads
26 March	Hillsong Church, Bella Vista 11 am–12.30 pm

Reference

Measles outbreak – April 2017 – Alerts. (2017). NSW.gov.au; NSW Health. https://www.health.nsw.gov.au/Infectious/alerts/Pages/april-measles.aspx

CASE STUDY 9.1.2

Measles outbreak originating from Disneyland, USA, December 2014

Figure 9.1.14 shows the confirmed cases of measles reported to the California Department of Public Health from a single original infection. All cases were traced back to the original infection in Disneyland on 16–21 December, in which rash onset was 16 December.

A total of 110 California residents were confirmed to have measles between 27 December and 8 February. Thirty-nine visited one or both Disney theme parks during 17–20 December, where it is thought that they were exposed to measles. Thirty-four are secondary cases from the first wave of 38 infections, while 37 have an unknown exposure source. Twenty-six of the 34 secondary cases were household or close contacts, and eight received the virus in a community setting. Five of the California patients reported being in one or both Disney theme parks during their exposure period outside of 17–20 December, but their source of infection is unknown.

Among this outbreak of the 110 patients, a total of 96 were either unvaccinated or had unknown or undocumented vaccination status for measles. Within this group, there were 12 infants who were too young to be vaccinated. The infected patients were of various ages, ranging from 6 weeks to 70 years.

Review

1. Provide a plausible model that shows three waves of infections for the outbreak from Disneyland in December 2014, including dates (or date ranges) to February 2015, using information from case studies 9.1.1 and 9.1.2.
2. Estimate the number of initial infectious individuals who may have caused the measles outbreak from Disneyland, assuming the reported R_0 of 15–16 by the CDC for measles.
3. Research the vaccination rate for measles in California and predict the spread of disease after 8 February.
4. Estimate the total number of exposed local individuals in Disneyland during 16–23 December, using the vaccination rate for measles in California.
5. Identify the limitations to estimating the number of individuals exposed in Question 4.

Confirmed cases of measles reported to the California Department of Public Health, December 2014 to February 2015
Number of confirmed measles cases
exposure at Disney theme park
December 2014
January 2015
February
Date of rash onset

FIGURE 9.1.14 The confirmed cases of measles reported to the California Department of Public Health from December 2014 to February 2015

Reference

Centre for Disease Control and Prevention. (2015). *Measles outbreak California December 2014–February 2015*. https://www.cdc.gov/mmwr/preview/mmwrhtml/mm6406a5.htm

CASE STUDY 9.1.3

MERS outbreak in South Korea originating in Saudi Arabia, June 2015

MERS (Middle East Respiratory Syndrome) is a viral respiratory illness caused by the coronavirus (Figure 9.1.15). Coronaviruses are common throughout the human population and most people contract them at some point. However, usually only mild to moderate flu-like symptoms develop (Figure 9.1.16). The MERS coronavirus is new to humans and was first discovered in 2012 in Saudi Arabia. Most people who contracted the MERS coronavirus developed a severe respiratory disease. However, a small number have mild or no symptoms.

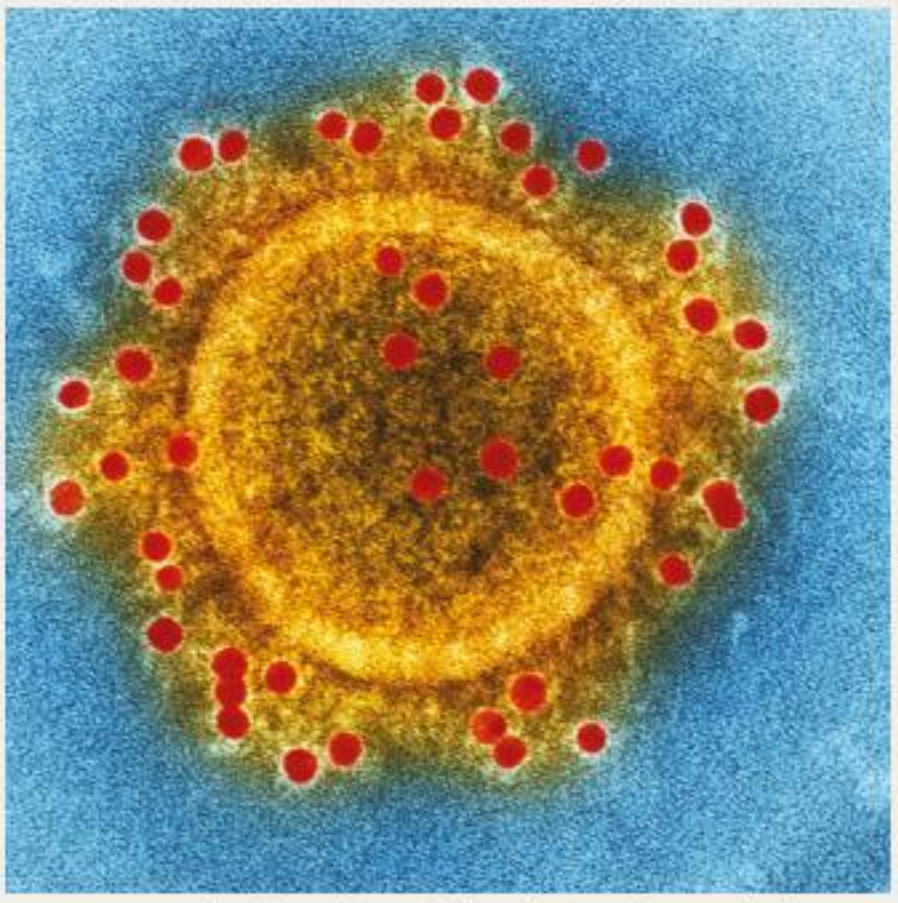


FIGURE 9.1.15 A coloured transmission electron micrograph of a MERS coronavirus (MERS-CoV) with antibodies (red spheres) attached to the viral envelope proteins.

An infected case arrived in Seoul, South Korea, and visited three locations, St. Mary's Hospital (Pyeongtaek), 365 Yeollin Clinic (Seoul) and Asan Seoul Clinic (also known as the Asan Medical Center, Seoul), shown in Figure 9.1.17. There was an outbreak of MERS at St. Mary's Hospital (Pyeongtaek), including greater than 34 secondary and tertiary infections. An individual from St. Mary's Hospital (Pyeongtaek) transmitted the infection to the Samsung Medical Center (Seoul) and the Goodmorning Hospital (Pyeongtaek). More than 60 infections were diagnosed at the Samsung Medical Center (Seoul). The first case of MERS was diagnosed in Seoul on 20 May, and the outbreak data was recorded on 12 June.

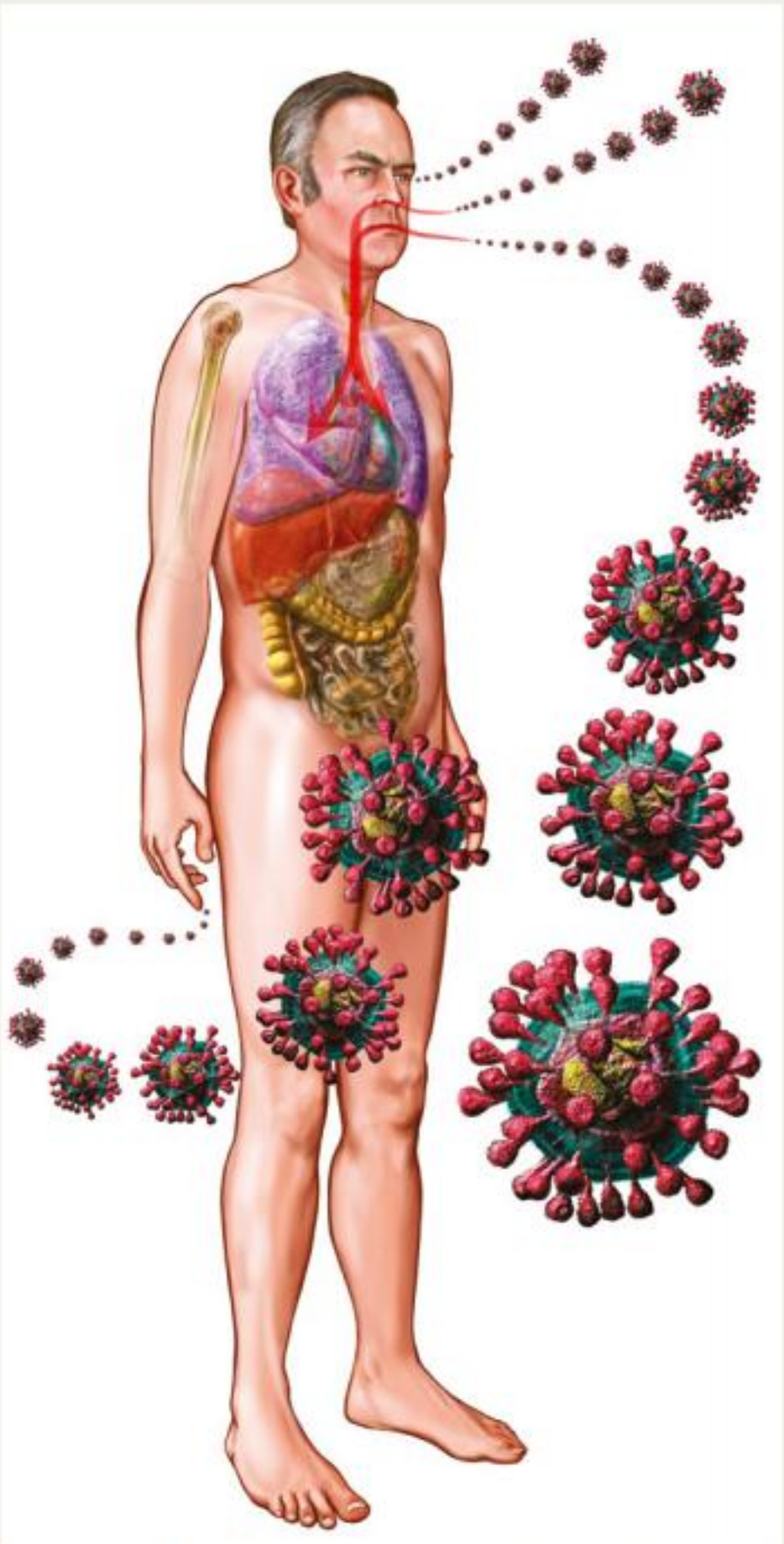


FIGURE 9.1.16 The MERS virus causes severe acute respiratory syndrome (SARS). It infects the lungs where it causes coughing, sneezing and watering eyes. This spreads the virus in liquid droplets through the air. It can also be contracted by touching contaminated surfaces or fluids.

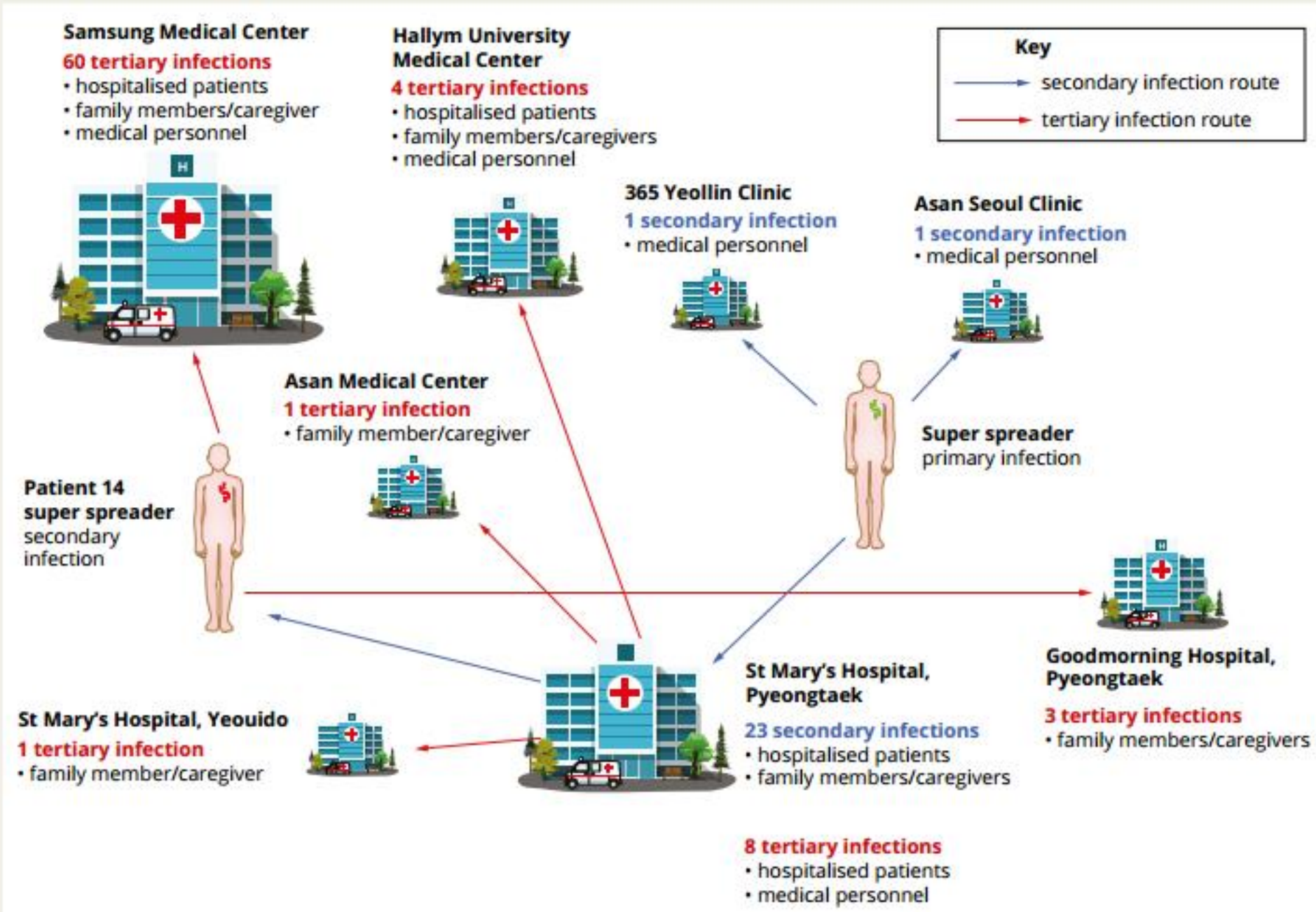


FIGURE 9.1.17 MERS outbreak in South Korea, June 2015. The data is represented as a schematic, showing the initial infectious individual (super spreader) in green, secondary infections in blue and tertiary infections in red.

Review

1 Specify the waves of infections from the evidence in Figure 9.1.17.

2 Provide a plausible reason, using your knowledge of adaptive immunity, for why the super spreader and Patient 14 only caused one outbreak each.

3 Suggest a mechanism of transmission supported by:
- sufficient and relevant evidence
- relevant patterns (Question 1 or 2)
- a justified scientific argument using concepts of mechanisms of transmission.

4 According to WHO and the CDC, the transmission of MERS is not well understood. Current understandings of MERS transmission are that it is transmitted through direct contact, including droplets from coughing, despite not being an airborne disease. Health authorities and disease researchers in Seoul, South Korea, who studied MERS during the outbreak believe their data contradicts current understandings.
Identify the data the WHO and CDC would have observed to determine MERS is not airborne.

5 Considering the information above, what would be a possible R_0 value for MERS and why?

Reference

World Health Organization. "MERS-CoV Outbreak Largest Outside Kingdom of Saudi Arabia." *Www.who.int*, World Health Organization, 2015, www.who.int/news/item/02-06-2015-mers-cov-outbreak-largest-outside-kingdom-of-saudi-arabia.

CASE STUDY 9.1.4

Influenza outbreak in Queensland, Australia, June 2017

In 2017, a new strain of the influenza virus spread across Australia, causing more hospitalisations and deaths than recorded previously. Figure 9.1.18 shows data from the Australian Government Health Department July report, which included data recorded up to 21 July. Before the next report was published, the Queensland Medical Association delivered updated data at a press conference on 11 August, stating that Queensland had recorded 14455 cases since 1 January 2017 and in the past week more than 3500 new cases had been identified

Review

1. Use Figure 9.1.18 to predict the number of influenza notifications in Queensland per week until week 52 of 2017.
2. What is a possible reason for an earlier peak of infected people in the Northern Territory compared to other states?

a

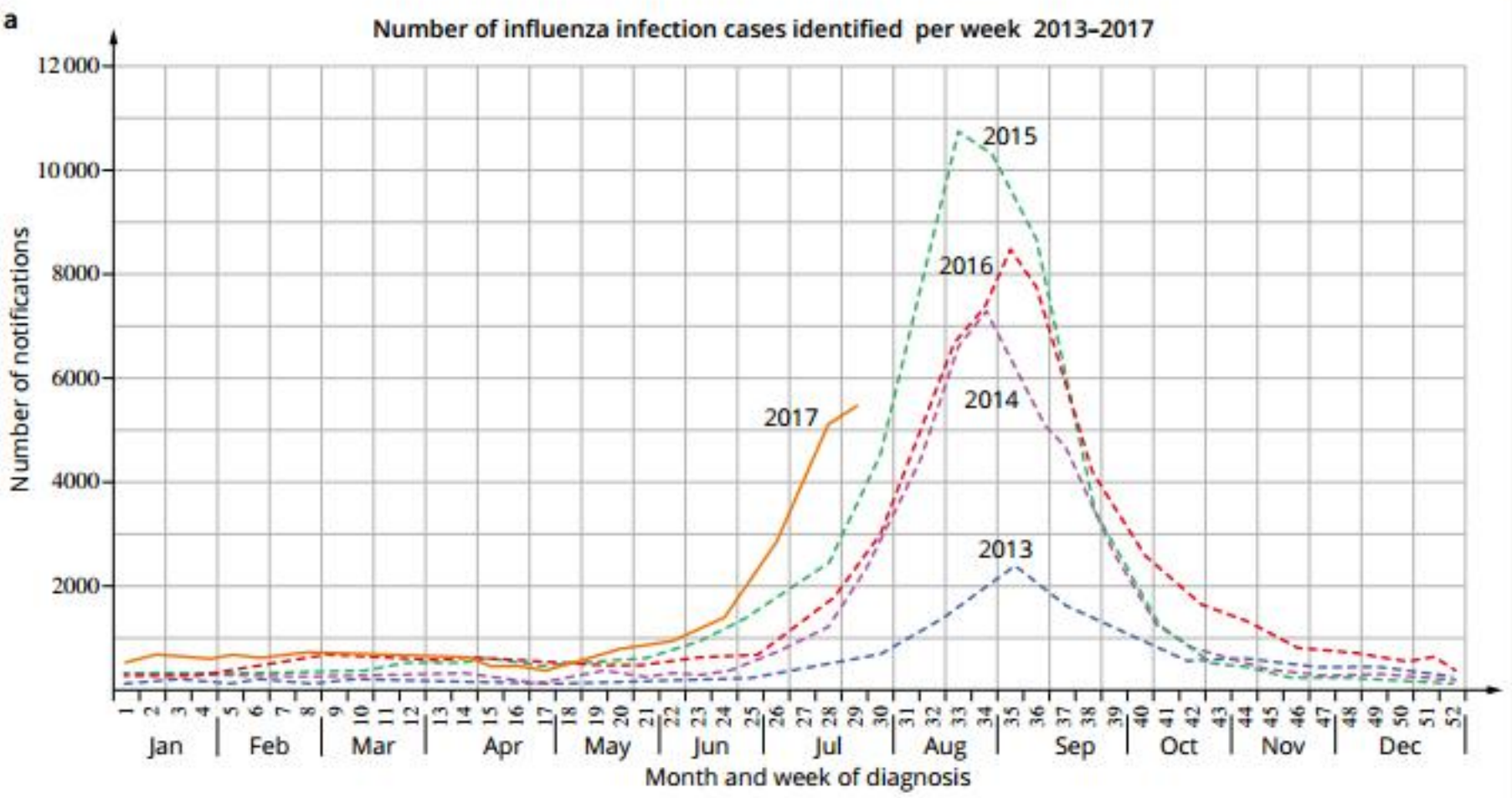
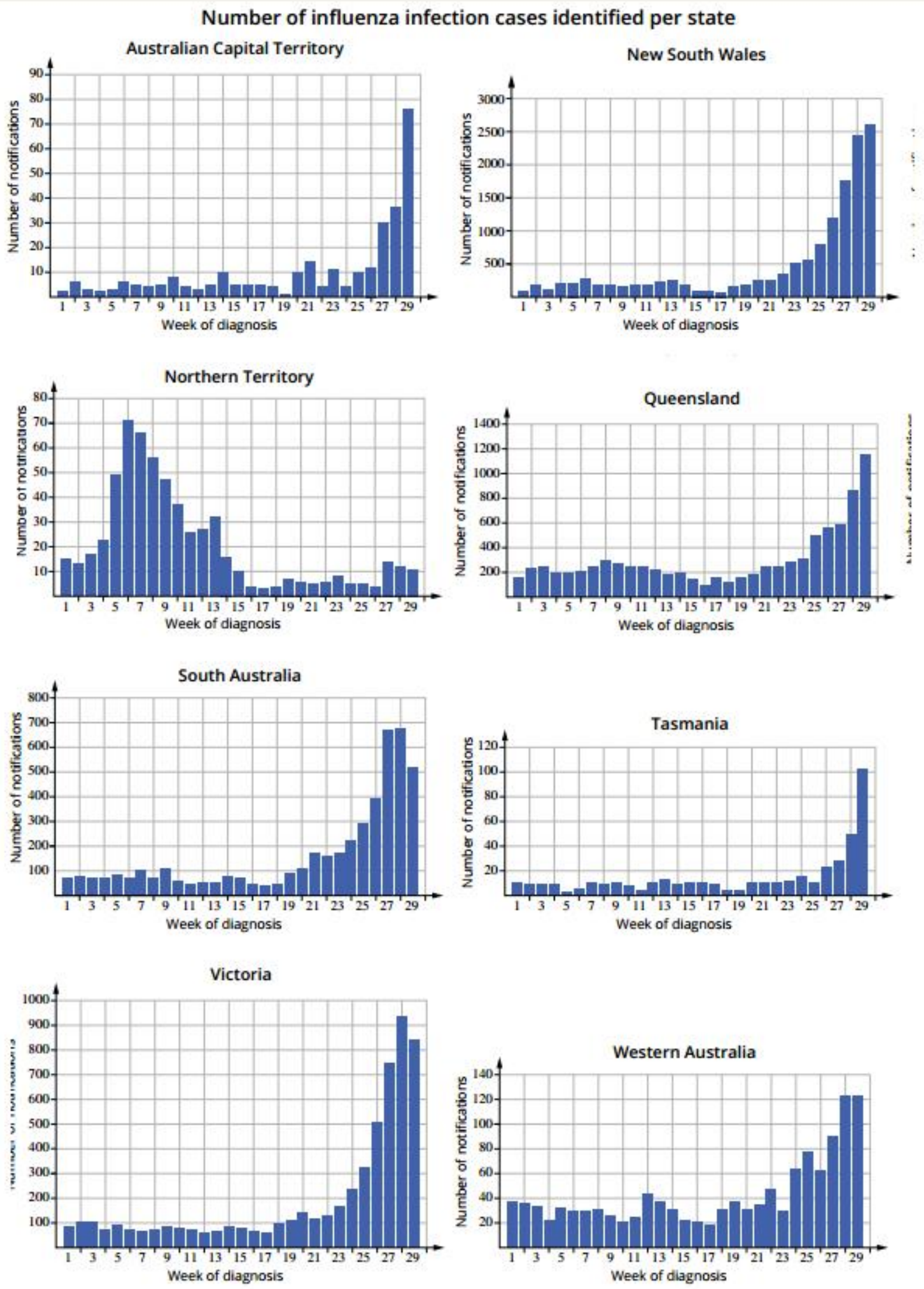


FIGURE 9.1.18 (a) Number of flu infection cases identified by health professionals from 2013 to 2017. (b) Number of flu infection cases identified per state by health professionals from 1 January to 21 July per week.

Reference

Mackay, I. M., & Arden, K. (2017). *Here's why the 2017 flu season was so bad*. University of Queensland Australia. https://medicine.uq.edu.au/article/2017/11/heres-why-2017-flu-season-was-so-bad

b


Number of influenza infection cases identified per state
Australian Capital Territory
Number of notifications
Week of diagnosis
New South Wales
Number of notifications
Week of diagnosis
Northern Territory
Number of notifications
Week of diagnosis
Queensland
Number of notifications
Week of diagnosis
South Australia
Number of notifications
Week of diagnosis
Tasmania
Number of notifications
Week of diagnosis
Victoria
Number of notifications
Week of diagnosis
Western Australia
Number of notifications
Week of diagnosis

CASE STUDY 9.1.5

Malaria epidemic in sub-Saharan Africa

Malaria is a disease caused by parasitic protozoans. It results in flu-like symptoms, fever, chills, headache and nausea. Left untreated, malaria can cause severe disease and death. WHO estimates that there were 188 million cases of infection in sub-Saharan Africa in 2015, comprising 88% of the world's total malaria cases (214 million). WHO also estimates that malaria caused 394 000 deaths in sub-Saharan Africa. WHO stated that, at the time of their report, nine sub-Saharan African countries had reliable estimates of infections from medical clinics and regional programs.

The parasitic protozoan that causes malaria undergoes many stages in its life cycle, which requires at least two different organisms—humans and female *Anopheles* mosquitoes (Figure 9.1.19). In humans, the juvenile protozoans grow and multiply in the liver, where they mature. They can lie dormant in the liver for weeks or even up to 4 years before continuing their life cycle. Eventually, they multiply in the infected liver cells until they rupture, and the mature protozoans are released into the bloodstream. The mature protozoans then multiply asexually within red blood cells until they rupture and release mature daughter protozoans. These daughter protozoans will either return to the liver to continue the cycle or they will circulate in the bloodstream until another *Anopheles* mosquito ingests them.

The cycle continues, they invade other red blood cells, and they increase in number. The mature protozoans also release male and female gametes into the blood.

While in the blood, the parasite causes symptoms such as anaemia, fever, chills, nausea, flu-like illness, coma and death. Also, while in the blood, the female *Anopheles* mosquito (Figure 9.1.20) receives the protozoan gametes when feeding, and the cycle continues.

After 10–18 days, the parasites are found (as sporozoites) in the mosquito's salivary glands. When the *Anopheles* mosquito has a blood meal from another human, the sporozoites are injected with the mosquito's saliva and start another human infection when they parasitise the liver cells.

Thus, the mosquito carries the disease from one human to another (acting as a vector). Unlike the human host, the mosquito vector does not suffer from the presence of the parasite.

Malaria is treatable with medication, but the medication is costly and may have side effects after long-term use. A vaccine for malaria is not currently available although preliminary human trials are being conducted on a few potential vaccines. There are a couple of complications related to dealing with the epidemic in sub-Saharan Africa.

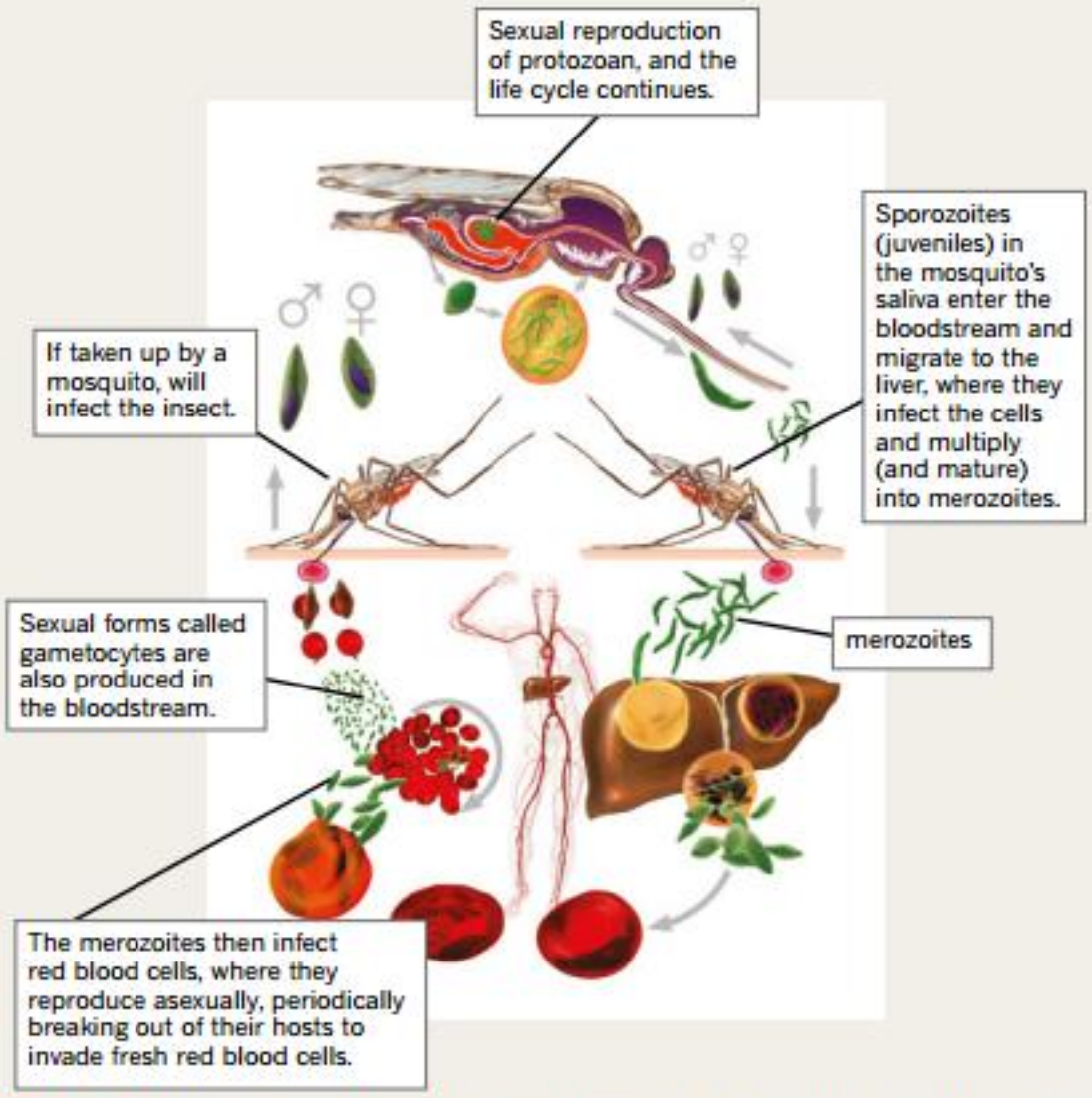

Sexual reproduction of protozoan, and the life cycle continues.
Sporozoites (juveniles) in the mosquito's saliva enter the bloodstream and migrate to the liver, where they infect the cells and multiply (and mature) into merozoites.
If taken up by a mosquito, will infect the insect.
merozoites
Sexual forms called gametocytes are also produced in the bloodstream.
The merozoites then infect red blood cells, where they reproduce asexually, periodically breaking out of their hosts to invade fresh red blood cells.

FIGURE 9.1.19 The protozoan life cycle is completed between mosquitoes and humans, which causes disease in humans.

FIGURE 9.1.20 A coloured scanning electron micrograph of *Anopheles gambiae*, female mosquito, one of the vectors of malaria.

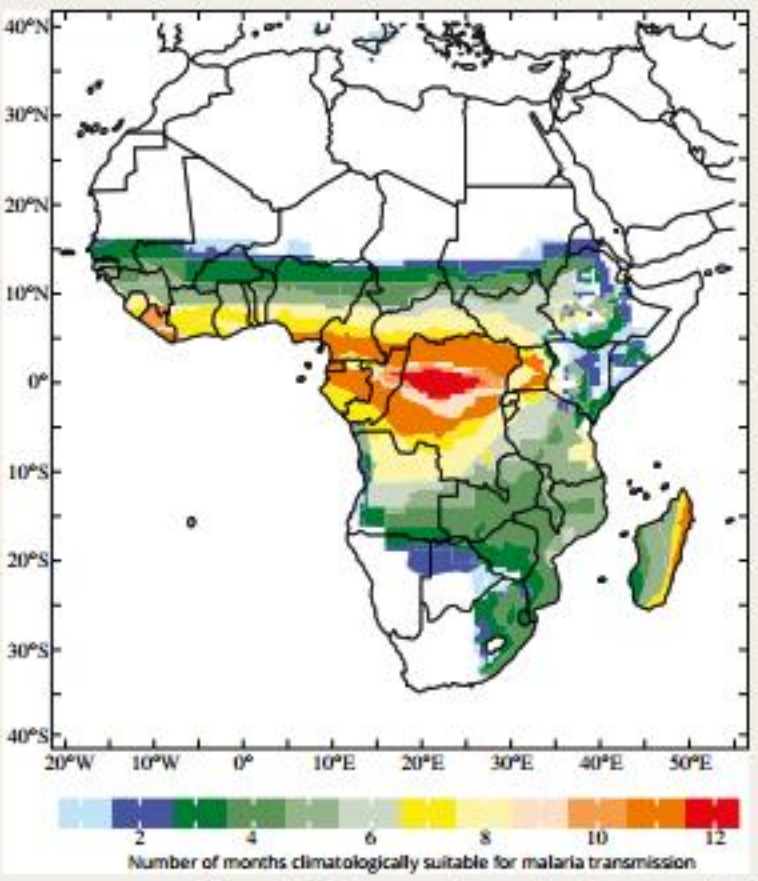


FIGURE 9.1.21 The sub-Saharan regions, showing the suitability to vector-borne malaria transmission due to climate. In general, the more central regions in the sub-Saharan region are more suitable to vector-borne malaria transmission.

Other complications include climate. The climate for approximately half the region is suitable for the mosquito vector to reproduce and grow, as illustrated in Figure 9.1.21. The female *Anopheles* mosquito requires water to lay her eggs. Conditions for mosquitoes to reproduce and grow can be temporarily suitable many times a year, depending on seasonal variations and rain. Other complications that hinder treatment include civil unrest, numerous governments, international borders and politics, as well as access to susceptible populations.

A recent strategy to decrease the number of infections (incidence) of malaria in the sub-Saharan region of Africa is an insecticide-treated mosquito net (ITN). In 2004, less than 4% of the sub-Saharan population slept under an ITN. This increased to approximately 50% in 2013, with 427 million nets delivered to the region since 2012.

Figure 9.1.22 shows the percentage of the sub-Saharan African population sleeping under ITNs from 2005 to 2013. Sub-regions still have little to no population sleeping under an ITN, while both the number of regions and the number of people sleeping under an ITN within regions is increasing. Some have reached 100%. Data in some countries has been validated by authorities, while others are estimates related to roll-out programs. Figure 9.1.23 shows the decrease of infections from 2005 to 2013 in general across the region.

Since 2010, many countries have reported that mosquitoes are developing resistance to the four ingredients used in an ITN. In 2000–2004, the resistance for each insecticide ingredient—carbamate, organochlorine, pyrethroid and organophosphate—was reported in approximately 3%, 21%, 19% and 4% of all countries worldwide, respectively. From 2010 to 2013, resistance was reported in 23%, 30%, 34% and 16% of all countries worldwide, respectively (Figure 9.1.24).

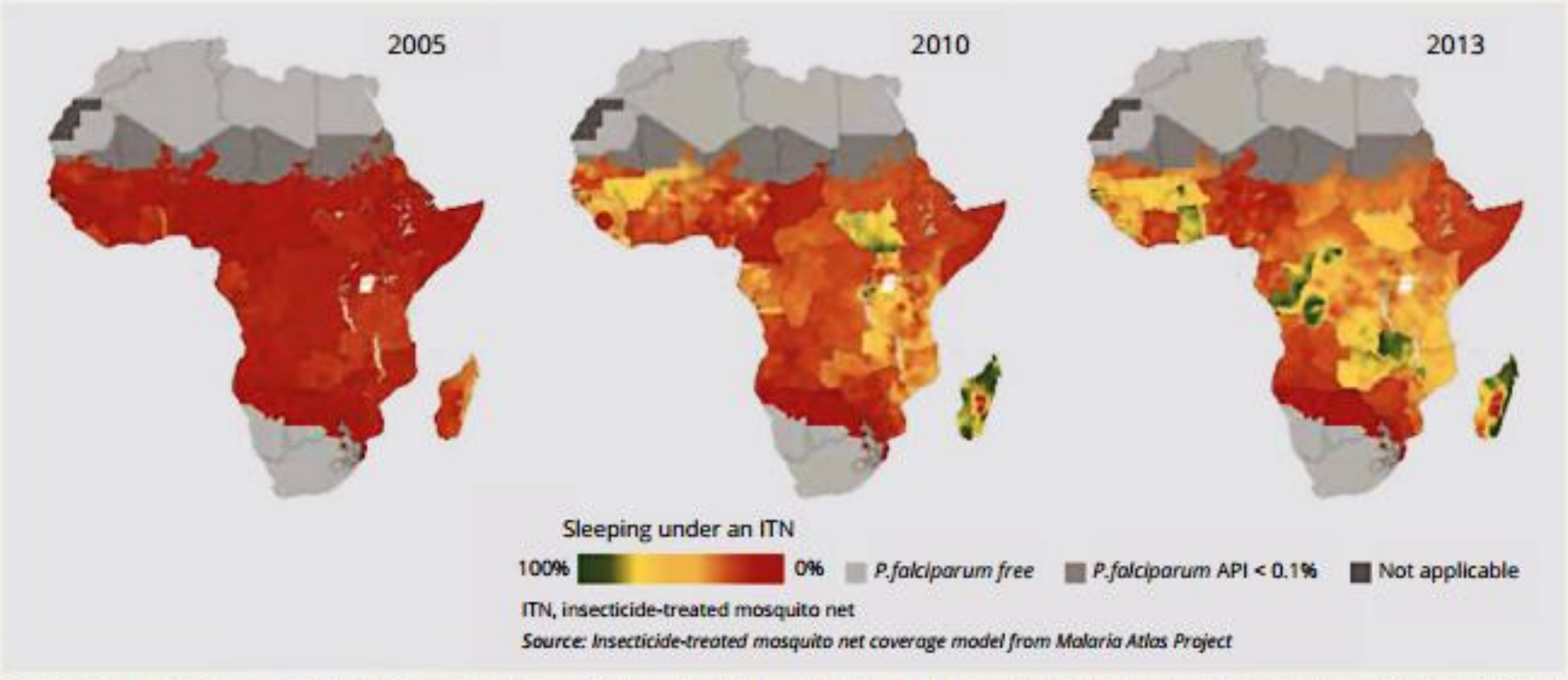


FIGURE 9.1.22 The percentage of the sub-Saharan African population sleeping under insect-treated bed nets has increased from 2005 to 2013.

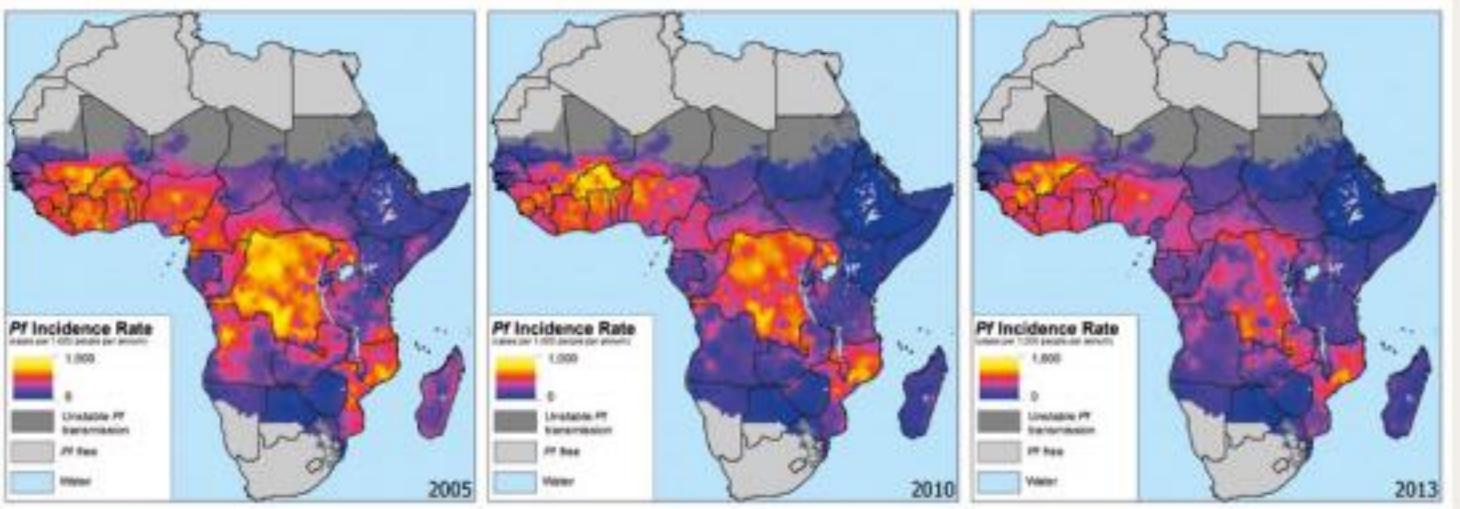


FIGURE 9.1.23 This incidence map has been developed from data estimating the incidence of vector-causing malaria across sub-Saharan Africa. It shows the decrease of infections from 2005 to 2013 in general across the region. Some areas still have exceptionally high numbers of infections.

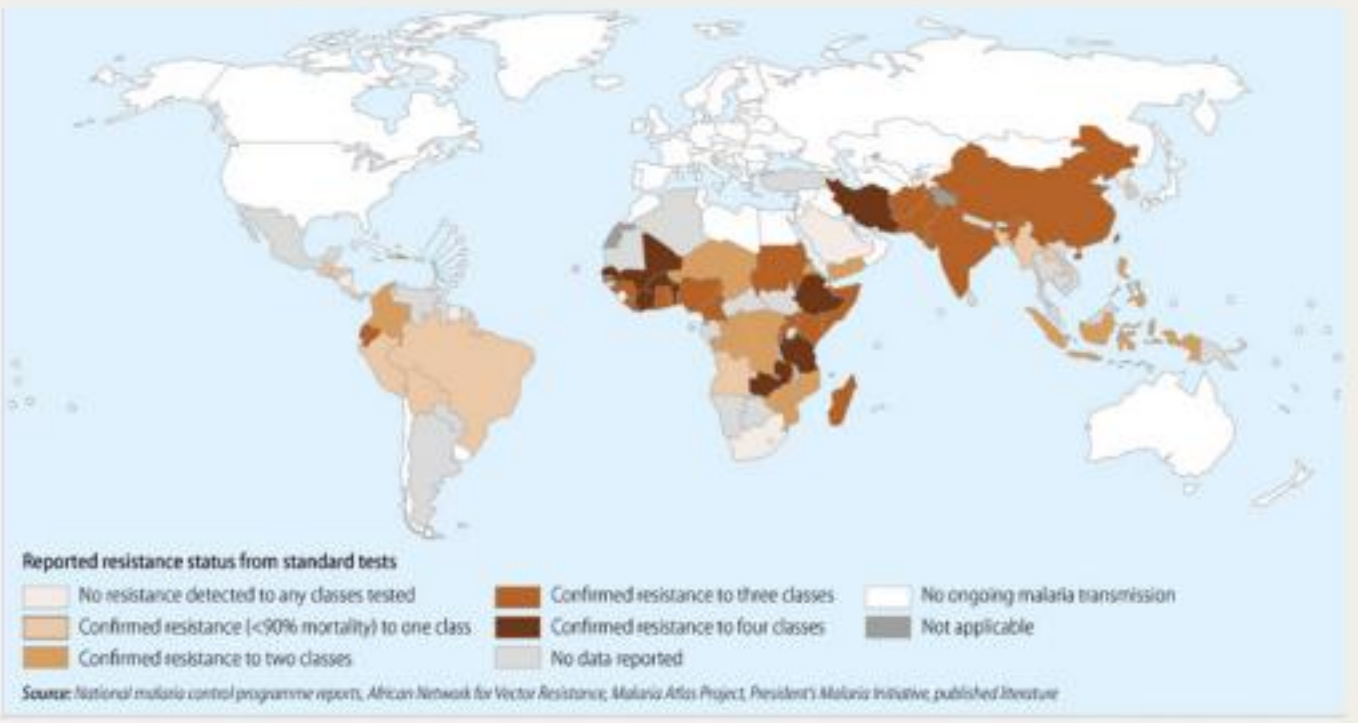

Reported resistance status from standard tests
No resistance detected to any classes tested
Confirmed resistance (<90% mortality) to one class
Confirmed resistance to two classes
Confirmed resistance to three classes
Confirmed resistance to four classes
No data reported
No ongoing malaria transmission
Not applicable
Source: National malaria control programme reports, African Network for Vector Resistance, Malaria Atlas Project, President's Malaria Initiative, published literature

FIGURE 9.1.24 The World Malaria Report 2014 illustrates the location and countries reporting insecticide resistance of ingredients found in ITNs.

Review

1. Determine whether or not the data supports a correlation between malaria and climate.
2. Identify the uncertainty for the data in Figure 9.1.21.
3. Explain the relationship between the use of ITNs across the sub-Saharan region and the incidence of malaria, specifying biological details that are relevant.
4. Identify limitations in the evidence regarding malarial infections and ITN use. Outline plausible effects of these limitations on the data in Figures 9.1.22 and 9.1.23.
5. Research and model the incidence of malaria since 2015 in sub-Saharan Africa.
6. Research and report on the effectiveness of ITNS in reducing incidences of malaria since 2015 and are there any impediments to its use?

References

Bertozzi-Villa, A., Bever, C. A., Koenker, H., Weiss, D. J., Vargas-Ruiz, C., Nandi, A. K., Gibson, H. S., Harris, J., Battle, K. E., Rumisha, S. F., Keddie, S., Amratia, P., Arambepola, R., Cameron, E., Chestnutt, E. G., Collins, E. L., Millar, J., Mishra, S., Rozier, J., & Symons, T. 2021, 'Maps and metrics of insecticide-treated net access, use, and nets-per-capita in Africa from 2000-20', *Nature Communications*, *12*(1). https://doi.org/10.1038/s41467-021-23707-7

CDC. 2019, *CDC - Malaria - Malaria Worldwide - How Can Malaria Cases and Deaths Be Reduced? - Insecticide-Treated Bed Nets*, CDC. https://www.cdc.gov/malaria/malaria_worldwide/reduction/itn.html

Cohee, L., & Laufer, M. 2018, 'Tackling malaria transmission in sub-Saharan Africa', *The Lancet Global Health*, *6*(6), e598–e599. https://doi.org/10.1016/s2214-109x(18)30197-9

Lindsay, S. W., Thomas, M. B., & Kleinschmidt, I. 2021, 'Threats to the effectiveness of insecticide-treated bednets for malaria control: thinking beyond insecticide resistance', *The Lancet Global Health*, *9*(9). https://doi.org/10.1016/s2214-109x(21)00216-3

CASE STUDY 9.1.6

Communicable diseases

Communicable diseases are diseases that are infectious from person-to-person, either directly, indirectly or through a vector. Across the globe, they are major causes of death and socioeconomic disruption to businesses and governments. For some countries, communicable diseases can result in economic instability. For other countries, communicable diseases can result in millions or billions of lost dollars for governments and major employers. It is almost inevitable that taxpayers will bear the cost of the financial impact of communicable diseases, to varying levels.

The Australian Department of Health requires medical practitioners to enter data into the National Notifiable Diseases Surveillance System to record all cases of communicable diseases in Australia. This is extremely important because it provides real data to health organisations and medical practitioners about current disease trends and communicable issues that affect their patients. Each fortnight, the surveillance system produces the *National Communicable Diseases Surveillance Report*. A summary of the report is shown in Table 9.1.10. With this data, the Australian Government can make informed decisions about appropriate vaccines, medications, early warnings and possibly earlier treatment to improve the health of individuals and communities. Planning and evaluation of disease prevention and control programs can be developed and, ultimately, reduce the spread of disease and the number of infections.

The data published in the *National Communicable Diseases Surveillance Report* allows for large-scale data analysis and longitudinal studies. Longitudinal studies investigate the same sample (e.g. population) over a long period of time, such as years or decades. This helps authorities understand the effects over years, regardless of the number of events and how these may influence other factors. The Australian Government can use the information to develop educational campaigns, evaluate health systems, inform the public and distribute funds where appropriate to help minimise the spread of disease in Australia.

Limitations of the *National Communicable Diseases Surveillance Report* include that it only records infectious people who visit a medical practitioner, and the number of infections for only the diagnosed diseases. Other limitations are that some communicable diseases are, or can be, **asymptomatic**, some can be a '24-hour bug', where people recover from acute illness within a day or two and do not seek medical assistance, while others are misdiagnosed as stress, anxiety or something other than a communicable disease.

TABLE 9.1.10 A summary of the year to date (YTD) reported communicable disease data in Australian states and territories by the Australian Government Department of Health from the *National Communicable Disease Surveillance Report* (18 July 2017)

Disease	ACT	NSW	NT	Qld	SA	Tas	Vic	WA	2017 YTD
hepatitis B (unspecified)	0	61	0	44	3	0	56	31	3471
hepatitis C (unspecified)	4	147	5	102	1	3	70	54	5963
campylobacteriosis (gastrointestinal)	15	–	18	262	127	31	44	120	12928
cryptosporidiosis (gastrointestinal)	0	18	2	22	12	1	39	7	3795
salmonellosis	8	81	11	104	28	7	98	83	11200
chlamydial infection	45	882	53	938	177	27	0	348	48488
gonococcal infection	9	276	36	161	45	3	221	118	16896
influenza (laboratory confirmed)	174	7182	24	2973	1363	194	1959	283	42521
pertussis	9	221	3	77	34	1	53	60	7279
varicella zoster (unspecified)*	10	–	0	449	1032	19	2	97	10258
varicella zoster (shingles)*	10	–	11	0	54	0	7	57	4440
Ross River virus infection	0	14	4	71	2	2	5	11	6348

*South Australia (SA) notified a retrospective varicella dataset, including data with diagnosis dates from 2012 to 2017.

The flu, caused by the influenza virus, is an example of where the National Notifiable Diseases Surveillance System records only a fraction of the infections across Australia each year. It is well accepted that only a portion of influenza infections are recorded each year. However, the longitudinal data enables predictions and trends to be known even with a limited number of recorded cases.

Another example is chlamydia, a bacterial infection of the reproductive and urinary systems. The 2016 *National Blood-borne Viruses and Sexually Transmissible Infections Surveillance and Monitoring Report* published by the University of New South Wales Kirby Institute estimated that there were 257 545 chlamydia infections in 15–29 year olds in Australia in 2015. The Notifiable Diseases Surveillance System only recorded 93 142 in total for Australia. The difference is attributed to 80% of people infected with chlamydia being asymptomatic.

Every year, there is a 'peak flu season' in which the number of influenza infections dramatically increases and peak before dramatically declining (see Case study 9.1.4). To combat the number of infections, illness, death and strain on the health system (e.g. use of beds, **quarantine** for the highly infectious, attention required by medical staff and governments' funds to nurse patients) due to influenza, one preventative action plan by the government is the annual flu vaccination. This vaccine is subsidised by the government every year for people in the high-risk category. Also, many workplaces offer the government flu shot (vaccine) free to their employees to minimise the number of days employees miss work due to influenza and thus minimise the loss of income to the business.

The difficulties and limitations for the influenza vaccine, and the preventative action plan, are that each year the peak flu season can be caused by a different viral strain. Health authorities state that international travel and antigenic drift and shift contribute to the annual changes in the strain. Often, Australians returning from international travel are the greatest source of the strains. Typically, the trend is a 12-month lag from either the USA or Western Europe. Antigenic shift is another major influence on the peak flu season. However, it is often too complex to understand the source.

Review

1. Use research to create a list of diseases from Table 9.1.10 that are treatable with medication.
2. Indicate possible reasons why people do not visit a medical practitioner when they are ill, resulting in unrecorded infections. How could this loss of data be overcome?
3. A database for the Australian Government Health Department, National Notifiable Disease Surveillance System is available at the following URL: https://nindss.health.gov.au/pbi-dashboard/. Scroll down to the bottom and click on the tab labelled 'Notifications for all diseases'. Using this page, determine a 10-year trend for the two most reported communicable diseases in Table 9.1.10 on page 389 and one other.
4. Develop an action plan, Medicare rebate policy or educational program to reduce the number of infections for one of the diseases in your response to Question 3. Ensure your action plan, Medicare rebate policy or educational program is justified by a rationale.

References

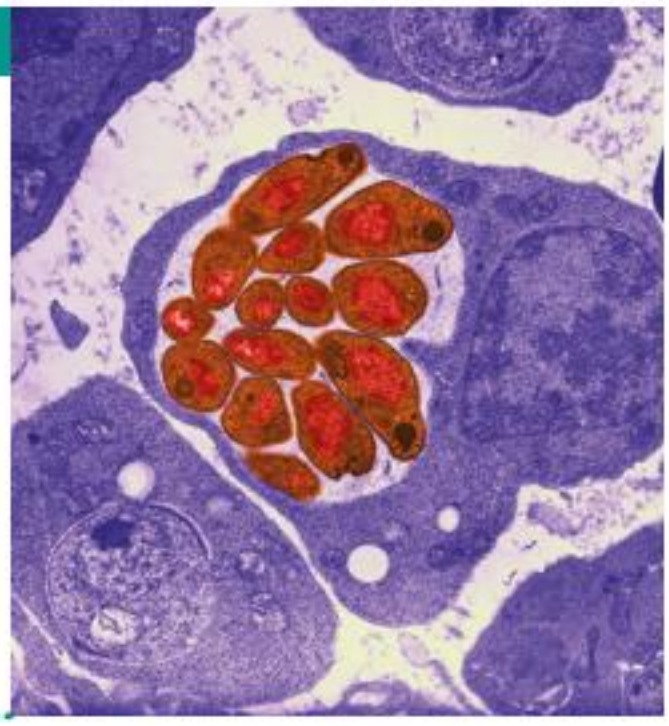
Department of Health and Aged Care. 2022, *Communicable diseases*, Australian Government Department of Health and Aged Care. https://www.health.gov.au/topics/communicable-diseases

Department of Health and Aged Care. 2023, *National Notifiable Diseases Surveillance System (NNDSS)*, Australian Government Department of Health and Aged Care. https://www.health.gov.au/our-work/nndss#data-reports

9.2 Epidemiology and the control of disease

BY THE END OF THIS MODULE, YOU SHOULD BE ABLE TO:

- explain the importance of personal hygiene measures in controlling the spread of disease including strategies to control the spread of disease at the community level, such as:
  - contact tracing and quarantine
  - school and workplace closures
  - reduction of mass gatherings
  - temperature screening
  - travel restrictions
- analyse data to predict outbreaks, their sources and modes of transmission and determine strategies to control their spread
- interpret data for the modelling of the spread of disease including long term immune response data.



In this module, you will learn measures and strategies of personal hygiene that minimise the spread of disease and transmission between individuals and throughout communities. You will learn about the effectiveness of hand washing, consider **hygiene etiquette** in disease transmission, and learn about the importance of hygiene etiquette while travelling. Community measures to control the spread of disease will be covered that relate to mass gatherings, quarantine strategies and temperature screening. Combating outbreaks requires considerable effort from many organisations and personnel. Modelling and preparing for potential outbreaks, or monitoring for source (initial) infections, can enable responses that protect communities.

PERSONAL HYGIENE TO CONTROL SPREAD OF DISEASE

Hygiene refers to actions that lead to cleanliness and good health. The spread of disease can be minimised and controlled through appropriate personal hygiene. Personal hygiene involves practices that an individual performs on his or her own body to maintain cleanliness, health and wellbeing. Examples of personal hygiene practices shown in Figure 9.2.1 include:

- washing hands, other body parts and hair with soap and clean water
- avoiding coughing and sneezing on others
- cleaning items that you come into contact with when you are ill
- appropriately disposing of tissues and other items that may contain germs
- using protective barriers, such as gloves and condoms (appropriately and consistently), when there is a risk of infection.

Clean running water is not always available in some countries, communities or locations and so achieving personal hygiene can sometimes be difficult. This can be the case in remote locations, locations beyond urban development where infrastructure does not exist, in underdeveloped countries and communities where poverty and famine are common, war-torn regions and refugee camps. If personal hygiene is not maintained, many diseases can spread.

FIGURE 9.2.1 (a) Washing hands removes microbes from skin. (b) Sneezing into a tissue minimises the spread of disease. (c) Cleaning commonly used areas is important after they have been used by someone with an infection. (d) Gloves provide a protective barrier to minimise any possible transfer.

1 Lather hands with soap.

2 Rub both palms together.

3 Rub each finger and between fingers.

4 Rub palms with finger nails.

5 Rub back of hand with finger nails.

6 Wash thoroughly and towel dry.

FIGURE 9.2.2 Washing hands correctly is an important step in preventing the spread of viruses or bacteria through contact. The CDC (Centre for Disease Control in the United States of America) recommends scrubbing hands for at least 20 seconds to ensure sufficient removal of any pathogens for disease prevention.

Hand washing

The transfer of pathogens by hands is the most common personal mechanism of pathogen transfer (see Figure 9.2.2). Some estimates state that 80% of infectious diseases involve transmission via hands. Generally, hand washing is seen as the primary preventative measure in disease transmission. For example, in the case of viral gastroenteritis, hand washing is more effective than options such as alcoholic hand sanitisers. Thorough washing with adequate quantities of water and plain soap removes 90% of transient (superficial) **contaminants** on the skin, including pathogens. To prevent the transmission of pathogens by hands, it is recommended you wash your hands:

- before and after eating or handling food or drink
- before providing medication
- after hands become visibly soiled
- after visiting the toilet
- after using a computer keyboard
- after handling laundry
- after handling waste
- after blowing, wiping or touching your nose or mouth
- after sneezing or coughing
- after contact with an ill person
- after contact with animals.

Disinfectants, alcohol-based soaps and hand sanitisers (with at least 60% alcohol) are highly effective **antibacterials** and in general kill 99.99% of microbes. However, these should only be used if water and soap is unavailable. Non-alcohol hand sanitisers may not work equally well for all microbes (e.g. gram-negative compared to gram-positive bacteria, and viral gastroenteritis), may develop resistance in the microbes, and may reduce microbe growth rather than kill them.

No single product will kill, remove or eliminate all microbes. It is important to use them appropriately. Alcohol-based sanitisers will not work effectively in too small volumes or if they are wiped off before they dry. They do not kill, eliminate or inactivate all microbes, and water and soap are more effective than sanitisers for some microbes. Research has also shown that alcohol-based sanitisers do not work well when hands are heavily soiled (visibly dirty) or greasy, such as after handling food, playing sports, working in the garden, camping or fishing.

CASE STUDY 9.2.1

Hand washing in schools, USA

Scientists in the USA investigated whether alcohol gel hand sanitiser could reduce the number of infections. The study sampled children from 16 schools in California, Delaware, Ohio and Tennessee. At each location, multiple districts were chosen and two schools in each district were selected as the sample population. Different locations and demographics were chosen, with schools being paired on the basis of similar population demographics and locations. Some information about the schools is listed in Table 9.2.1. Within each district, one of the paired schools was assigned as the control group (no alcohol gel hand sanitiser) and the other school was assigned as the test group or product group (alcohol gel hand sanitiser was used). A total of 6080 students were included in the study.

TABLE 9.2.1 Geographic and demographic characteristics of US school districts chosen to test the effectiveness of alcohol gel hand sanitiser

Location	Population	Per capita income	Classification of area
Cuyahoga Falls, Ohio	49913	$40700	suburban
Hudson, Ohio	22287	$26545	suburban
Athens, Tennessee	13340	$17512	rural
Irvine, California	127220	$28037	urban
Wilmington, Delaware	71500	$18011	urban

The study was conducted over a full academic year, of which the test groups were instructed to wash their hands with the alcohol-gel hand sanitiser when entering and leaving the classroom and after sneezing and coughing.

Absenteeism in the study was defined as the total number of school days not attended because of colds, flu and gastrointestinal disease. All other types of absenteeism were excluded. Absenteeism records were kept by school staff, who identified the reason for the absence. The schools provided the study coordinators with the records on absenteeism due to illness.

Table 9.2.2 shows the results of the study. The results showed less absenteeism in the group using the hand wash compared to the control group that did not.

Review

1. Identify and explain the valid methodologies employed by the hand washing study in US schools.
2. Explain the limitations of the study design when considering the selection of schools.
3. Explain the limitations of the sampling methodologies.
4. Determine how the percentage difference over control is calculated and infer what may have affected the calculations slightly.
5. Interpret the percentage difference over control.
6. Consider the appropriateness of percentage difference over control for the different sample sizes between Wilmington, Delaware, and Cuyahoga Falls, Ohio.
7. Outline how a similar experiment could be conducted in Queensland? What would be needed to insure the validity and reproducibility of your experiment.

Reference

Hammond, B., Ali, Y., Fendler, E., Dolan, M., & Donovan, S. 2000, 'Effect of hand sanitizer use on elementary school absenteeism', *American Journal of Infection Control*, *28*(5), 340–346. https://doi.org/10.1067/mic.2000.107276

TABLE 9.2.2 The results of a study on the effect of an alcohol gel hand sanitiser in schools

School district	Number of students	Grades	Students in product group	Students in control group	Days absent hand sanitiser group*	Days absent control group*	Days absent per student in hand sanitiser group	Days absent per student in control group	Per cent difference over control
Cuyahoga Falls, Ohio	2576	K–5	1440	1136	2327.5	2738	1.62	2.41	32.96†
Hudson, Ohio	818	2, 3	266	552	724	1697	2.72	3.07	11.49†
Wilmington, Delaware	223	3, 4	110	113	478	533	4.35	4.72	7.87
Athens, Tennessee	1272	K–6	680	592	1953	2102	2.87	3.55	19.07†
Irvine, California	1191	K–5	579	612	1959	1996	3.38	3.26	–3.75
Overall	6080	K–6	3075	3005	7441.5	9066	2.42	3.02	19.76†

* Numbers were normalised for equal populations before calculating percentage difference over control.
† Statistical significance ($P < 0.05$).

CASE STUDY 9.2.2

TABLE 9.2.3 Geographic and demographic characteristics of New Zealand school districts chosen to test the effectiveness of alcohol-gel hand sanitiser

Category	Characteristic	Hand sanitiser group		Control group	
		Number or mean	Percentage or SD	Number or mean	Percentage or SD
Schools	Total schools	34		34	
	roll just prior to study (mean, SD)	228.8	115.8	209.6	102.2
	City				
	Christchurch	19	55.9%	18	52.9%
	Dunedin	11	32.4%	12	35.3%
	Invercargill	4	11.8%	4	11.8%
	School decile*				
	1–3 (least advantaged)	5	14.7%	11	32.4%
	4–7	9	26.5%	6	17.6%
	8–10	20	58.8*	17	50.0%
	Total follow-up children	1301		1142	
Follow-up children	follow-up children with a baseline questionnaire	1287		1132	
	Household income ($A)				
	not stated	101	7.9%	87	7.7%
	0–40 000	156	12.1%	163	14.4%
	40001–80000	525	40.8%	415	36.7%
	>80 000	505	39.2%	467	41.3%
	Ethnicity (prioritised)†				
	Māori	163	12.7%	132	11.7%
	Pacific	31	2.4%	35	3.1%
	Asian	41	3.2%	33	2.9%
	European	1019	79.2%	907	80.1%
	other	25	1.9%	21	1.9%
	not stated	8	0.6%	4	0.4%
	Education (highest qualification in household)				
	no qualifications stated	51	4.0%	61	5.4%
	some high school qualification	334	26.0%	309	27.3%
	university	632	49.1%	515	45.5%
	alternative qualification	270	21.0%	247	21.8%
	Self-reported overall family hand hygiene				
	not stated	82	6.4%	58	5.1%
	poor/fair	115	8.9%	88	7.8%
	good/very good/excellent	1090	84.7%	986	87.1%
	number of people in household	4.4	1.12	4.36	1.07
	number of children in household	2.42	0.96	2.43	0.93
	Children under 5 years in household				
	0	903	70.2%	807	71.3%
	1	322	25.0%	275	24.3%
	2	58	4.5%	48	4.2%
	3	4	0.3%	2	0.2%
	Caregivers in paid employment				
	both caregivers, at least 20 h/week	514	39.9%	455	40.2%
	at least one caregiver <20 h/week	289	22.5%	219	19.4%
	at least one caregiver not in paid employment	478	37.1%	446	39.4%
	missing	6	0.5%	12	1.1%

SD = Standard deviation

† Respondents were asked to tick all the ethnicities represented in their household. Prioritised ethnicity, in New Zealand, codes as Māori participants who report Māori as one of their ethnic groups, as Pacific those who do not report Māori but do report a Pacific ethnicity as one of their ethnic groups, as Asian those who do not report Māori or Pacific but report an Asian ethnicity, and the remainder as European (if New Zealand European or another European ethnicity reported) or other (if not).

Hand washing in schools, New Zealand

A 2010 study in New Zealand investigated if the use of hand sanitiser of at least 60% alcohol in primary schools could reduce absenteeism due to illness. A total of 68 primary schools from the South Island within the areas of Canterbury, Otago and Southland were selected. Each school had to have at least 100 children and have no previous use of hand sanitisers. The schools in each area were randomly chosen to be a hand sanitiser group or control group, with equal representation of each (one of the researchers randomly assigned schools to each group without knowing which would be the hand sanitiser group or control group). A total of 50 students per school were randomly chosen, with a total of 2443 students involved in the study.

Table 9.2.3 shows the characteristics of participant school's families and students.

All schools received a single health education session on hand hygiene during term 1 (March and April) 2009. The study began in term 2 and concluded in term 3 (being conducted during the winter season of 27 April to 25 September 2009), with the hand sanitiser group applying 60% alcohol-based hand sanitiser during this time. The health education session ensured both groups had equivalent knowledge of hand hygiene and cleaning. The students were able to use the sanitiser at any time, but teachers ensured that the students used the hand sanitiser after coughing, sneezing and blowing their nose, and when leaving the classroom for morning break and lunch break.

The study measured the number of absence episodes due to any illness excluding injuries and infestation (head lice and scabies). A new absence was defined as an absence after at least 3 days without absence. The study collected the absence information from each school weekly. Results can be seen in Table 9.2.4. The ICC (interclass correlation coefficient) gives a measurement of reliability for the study, with a value of one being completely reliable and zero being unreliable. The *P* value, also called the probability value, is used to describe how likely it is that the data occurred by random chance. A *P* value of >0.05 indicates that there is no significant difference between the control group and the test group, and it is likely that the results occurred by chance. As with the ICC, the smaller the *P* value the less likely the results were obtained by random chance. In Table 9.2.4, the *P* value is 0.346 (>0.05), which indicates that there is no significant difference between the control group and the test group (hand sanitiser group).

Review

1. Identify and explain the valid methodologies employed by the hand washing study in New Zealand schools. Consider the locations, random association of groups, number of schools and previous use of sanitisers.
2. Explain the limitations of the design of the study when considering the selection of schools.
3. Explain the appropriate methodologies employed to choose the population sample. Consider sample size and sample selection of students.
4. Compare and contrast the reported results of the US study (Case study 9.2.1) and the New Zealand study (Case study 9.2.2) of hand washing in schools.
5. Explain whether or not it is valid to compare the results between the studies in Case study 9.2.1 and Case study 9.2.2.

Reference

Priest, P., McKenzie, J. E., Audas, R., Poore, M., Brunton, C., & Reeves, L. 2014, 'Hand Sanitiser Provision for Reducing Illness Absences in Primary School Children: A Cluster Randomised Trial', *PLoS Medicine*, *11*(8), e1001700. https://doi.org/10.1371/journal.pmed.1001700

TABLE 9.2.4 Results from New Zealand hand sanitiser study

Outcome	Number of children (34 schools per group)		Control group		Hand sanitiser group		IRR, hand sanitiser vs control (95% CI)	*P* value	ICC* (95% CI)
	Control group	Hand sanitiser group	Number of events (child days of follow-up)	Rate (per 100 child days or %)	Number of events (child days of follow-up)	Rate (per 100 child days or %)			
Primary outcome									
no. absences due to illness	1142	1401	1291 (111 451)	1.16	1542 (127 471)	1.21	1.06 (0.94, 1.18)	0.346	0.018 (0.012, 0.043)

Table 9.2.3 footnote:
* School level deprivation uses the decile assigned to each school by the New Zealand Ministry of Education for funding purposes. It reflects the proportion of students who live in more, or less advantaged communities, using information from the census on household income, occupation, household crowding, educational qualifications and income support. Decile 1 schools are in the least advantaged communities and decile 10 schools are in the most advantaged.

Hygiene etiquette

FIGURE 9.2.3 Hygiene etiquette is extremely important in medical practices to minimise transmission to susceptible individuals.

Good hygiene etiquette can interrupt the spread of disease, especially when an individual is sick. It can be exceptionally helpful to exercise hygiene etiquette when coughing or sneezing symptoms exist with an illness. Hygiene etiquette can minimise the spread of respiratory diseases that are highly infectious and cause a high number of infections and severe disease, such as influenza, respiratory syncytial virus (RSV), whooping cough and severe acute respiratory syndrome (SARS).

Coughing or sneezing into your hands or having unclean hands and then touching your face or other objects spreads disease to yourself and others. In this case, hygiene etiquette includes:

- covering your mouth and nose with a tissue when coughing or sneezing
- placing your used tissue in a rubbish bin
- washing your hands after you have coughed or sneezed
- coughing or sneezing into your elbow or the upper sleeve of your shirt if you do not have a tissue.

Hygiene etiquette in medical practices is especially important, as seen in Figure 9.2.3. Patients are exposed during invasive medical procedures, so medical staff must practice hygiene etiquette to minimise transmission directly into the body of a patient. In such cases, infection can take hold very quickly. Places of medical assistance and health services are where highly susceptible populations are located. Many people seeking assistance have compromised immune systems. Also, because ill and infected people congregate in these locations, there are high levels of infectious agents and pathogens. For these reasons, medical staff need to practice hygiene etiquette to minimise transmission of disease from one person to another.

Facemasks are most often used in Japan by people who have colds, so as not to infect others, or to avoid catching infections themselves (Figure 9.2.4). This practice has also been adopted by people in many other countries when experiencing illness, particularly during the recent COVID–19 pandemic.

FIGURE 9.2.4 In Japan, it is common to wear facemasks while commuting to work.

CASE STUDY 9.2.3

Coughing and sneezing etiquette

The Australian Government National Health and Medical Research Council has released the *Australian Guidelines for the Prevention and Control of Infection in Healthcare* (2019). Community groups and businesses can use this information to develop policies or inform the public. The 2019 report includes Guideline 3.1.5 Respiratory Hygiene and Cough Etiquette, which outlines cough etiquette, as shown in Figure 9.2.5.

Steps in respiratory hygiene and cough etiquette

- Cover the nose or mouth with disposable single-use tissues when coughing, sneezing, wiping and blowing noses.
- Use tissues to contain respiratory secretions.
- Dispose of tissues in the nearest waste receptacle or bin after use.
- If no tissues are available, cough or sneeze into the inner elbow rather than the hand.
- Practice hand hygiene after contact with respiratory secretions and contaminated objects or materials.
- Keep contaminated hands away from the mucous membranes of the eyes and nose.

* In healthcare facilities, patients with symptoms of respiratory infections should sit as far away from others as possible. If available, healthcare facilities may place these patients in a separate area while waiting for care.

FIGURE 9.2.5 The recommended etiquette for coughing outlined by the Australian Government National Health and Medical Research Centre

The 2019 *Australian Guidelines for the Prevention and Control of Infection in Healthcare* have been adopted by many businesses. Businesses can buy posters that inform employees about infection control. An example of a poster is in Figure 9.2.6.

COVER YOUR
COUGH AND SNEEZE
THE RIGHT WAY
DO: Use a tissue and place it immediately in the bin. Wash your hands or use a hand sanitiser.
DO: Use your upper sleeve. Germs won't spread through your clothing.
DON'T: Cover your cough with your hands. You'll end up spreading more germs via everything you touch.
FRESH & CLEAN

FIGURE 9.2.6 A workplace poster that details hygiene etiquette for coughing and sneezing

Review

1. Describe how the 2019 *Australian Guidelines for the Prevention and Control of Infection in Healthcare* would affect the transmission of influenza.
2. Compare the workplace poster in Figure 9.2.6 with the 2019 *Australian Guidelines for the Prevention and Control of Infection in Healthcare.*
3. Predict with justification which guidelines would most effectively decrease the spread of disease.

Reference

ACSQHC. 2019, Australian Guidelines for the Prevention and Control of Infection in Healthcare | Australian Commission on Safety and Quality in Health Care, www.safetyandquality.gov.au. https://www.safetyandquality.gov.au/publications-and-resources/resource-library/australian-guidelines-prevention-and-control-infection-healthcare

CASE STUDY 9.2.4

Travellers visiting Australia—Ross River virus

Queensland Health, a department of the Queensland Government, reports approximately 3000 cases of Ross River fever annually with occasional peaks of 5000–6000. The annual cases are generally dispersed evenly across the Queensland coast where water environments are close to larger populations. Areas of higher prevalence of Ross River fever include Cairns, Townsville, Central Queensland, Wide Bay, Sunshine Coast, Brisbane and the Gold Coast (including the hinterlands associated with each region).

Cases in Australia are correlated to climate and seasons. Tropical and subtropical regions of Australia report cases all year round (mostly in Queensland and Northern Territory), while southern parts of Australia (i.e. New South Wales, Victoria, southern South Australia and Tasmania) report seasonal infections.

Cases of Ross River fever in Europe are very rare and are typically found in individuals who are returning from Australia or the Oceanic region.

The Queensland Government provides information about Ross River virus on their website to inform locals and travellers about the symptoms, treatment and prevention of Ross River fever. The following information is a summary from the Queensland Government's website.

Ross River fever occurs widely in Australia. In Queensland it can occur throughout the year but it is most common between February and May.

The majority of people will recover within six weeks but in some individuals, the virus is prolonged. Symptoms occur within 3–11 days and include inflammation and pain in the joints as well as a fever followed by a raised red rash over the trunk of the body and limbs, which can last between one and ten days. Some individuals will be asymptomatic.

Unlike many other viruses it cannot be passed between humans and is only spread through mosquito vectors. The main reservoir hosts for the virus are animals such as kangaroos, wallabies, and possibly foxes, horses and some bird species. There are many species of mosquito that can act as a vector for Ross River virus and as there is no specific treatment for it, prevention is the most important precaution.

Specific recommendations include, avoiding outdoor areas during times of heavy mosquito infestations, using insect repellents and screening living and sleeping areas. Ensuring breeding areas such as pools of water are cleared from around your home or living space.

Review

A couple from the Netherlands are about to visit Australia and will spend 2 weeks in Queensland, 1 week in Cairns and 1 week on the Gold Coast. In Cairns, they plan to snorkel, visit the forest in the hinterlands, and swim in local swimming holes (rivers and waterfalls). In both destinations, they will spend a few days walking along trails in the hinterlands, including a visit to the glow-worms. On the Gold Coast, they will also visit the Currumbin Wildlife Sanctuary.

1. Provide advice the couple would be given by a travel doctor regarding their health in Australia.
2. Explain how the couple can avoid Ross River fever while coming into contact with kangaroos, horses, sheep and koalas at the Currumbin Wildlife Sanctuary.
3. Predict the potential for outbreaks of Ross River fever in infected travellers returning to their home in the following scenarios:
   - one couple returning to their home on the Nieuwe Maas, Rotterdam, Netherlands, in January
   - another couple returning to their home in Alto da Boa Vista, Rio de Janeiro, Brazil, in January.
4. Unlike Queensland, the Northern Territory and Western Australia, cases of Ross River fever are considered to be seasonal in New South Wales and Victoria. Why would this occur?
5. Mosquito eradication programs are considered the most effective way to reduce Ross River infections to humans. Why is this?

Reference

Queensland Government. 2017, Ross river virus – Viral infections, *Queensland Government*. https://www.qld.gov.au/health/condition/infections-and-parasites/viral-infections/ross-river-virus

TRAVEL HYGIENE

Many factors can influence a traveller's health as well as determine whether or not they develop disease from infections. One factor is exposure to a species or strain of a microbe that an individual may not have been exposed to before. This makes them susceptible to disease. Also, in foreign locations it is possible that different vectors and fomites exist that can transmit pathogens without travellers being aware.

When travelling to a new destination, you may need a different hygiene etiquette to maintain health. Therefore, it is especially important to develop an understanding of common vectors and fomites at foreign destinations when travelling and learn the local hygiene etiquette. The list of common recommendations below outlines typical hygiene etiquette when travelling to foreign destinations.

Developing the hygiene etiquette of local residents in a community is difficult. This is because local and expert knowledge is vast and sometimes not obvious. Also, expert knowledge continually develops, making local practices outdated. There may also be inconsistencies between traditional and up-to-date practices, making it difficult for a traveller to understand hygiene etiquette.

Before travelling, you should seek medical advice about diseases at your travel destinations. Health and disease organisations continually update their databases and information, improving understanding of disease transmission. This is valuable information for travellers. They can be informed about regions or areas to avoid, recommended vaccinations (Figure 9.2.7a) to receive prior to departing, or methods to minimise disease transmission (e.g. vector control using insecticides or repellents), as seen in Figure 9.2.7b. They can also learn about hygiene etiquette for common diseases, typical fomites and other mechanisms of transmission, all of which enable travellers to plan or develop strategies to maintain good health.

FIGURE 9.2.7 (a) Receiving vaccinations specific to the destinations of international travel. (b) Insect repellents are a valuable product to have when travelling. They minimise the incidence of infections that are transmitted by insect vectors.

FIGURE 9.2.8 The rapid bubbling of the water in a rolling boil, which is greater than when simmering.

Common recommendations to minimise infections and disease transmission while travelling include the following. Most relate to the individual or water.

- Drink only bottled water.
- Do not use tap water to clean your teeth.
- If no clean or other water source is available, boil water for 1 minute at a rolling boil before drinking it (Figure 9.2.8).
- When washing your hands, ensure they are totally dry before touching food.
- Do not wash fruit or vegetables in unsafe water.
- Ensure that dishes, cups or any other utensils are completely dry after they are washed.

COMMUNITY MEASURES TO CONTROL SPREAD OF DISEASE

Identifying infections and controlling the spread of disease at the community level is difficult. All the factors and variables related to controlling the spread of disease in individuals are compounded at the community level. The technology and strategies that can reveal disease or infections have been developed for individual detection. Currently, community detection does not exist. Therefore, the measures implemented to control the spread of disease through communities are based on modelling and predictions.

Contact tracing enables health authorities to monitor, provide and prioritise health care. This strategy uses the resources for disease detection and prevention at the source in the hope of eradicating transmission any further. Mass gatherings, which are unique, place immense strain on health authorities and resources. They require imperative planning and preparation. Temperature screening can test multiple individuals in a community or large samples of a population quickly. It is usually used at borders to respond rapidly and protect the larger population of cities and countries from new pathogens. Quarantine is the most effective strategy, and one of the oldest ways of combating the spread of disease. By isolating the ill and infected, disease cannot spread.

Contact tracing

Contact tracing is a strategy to identify and monitor infected individuals. Once an individual has been confirmed as infected, they are questioned about their activities as well as the activities, responsibilities and occupations of other individuals who have been around them since the onset of the illness. This includes identifying individuals through direct and indirect contact, and could include family members, work colleagues, friends, healthcare providers and service staff in the hospitality industry.

The process of contact tracing lists all individuals considered to have had contact with the infected person. Then, attempts are made to identify every listed individual so that they can be informed of their contact status with the infected individual. They are monitored, their symptoms are surveyed, and early care is provided if necessary. Contact tracing continues with a list of possible contacts for secondary infections. In some cases, high-risk contacts are quarantined or isolated, either at home or in hospital.

The process of contact tracing was employed for case studies 9.1.1, 9.1.2 and 9.1.3 in Module 9.1 and the value of tracing back to the origin of infection is evident. It can find individuals in the community who are possibly infected, provide early care and minimise the spread of disease, and enable the determination of R_0. It is a commonly used strategy in disease **epidemiology**. It is one of the only strategies that can provide information for diseases that are asymptomatic, lie dormant or have long incubation periods. Examples of diseases with long incubation periods are infectious mononucleosis (glandular fever) and hepatitis B. Chlamydia is most often asymptomatic, and shingles (herpes zoster) can lie dormant for decades.

When outbreaks occur, contact tracing is one of the first strategies employed. Examples are the case studies in Module 9.1, the Zika virus in South America, bird flu and swine flu in Asia, Ebola (Figure 9.2.9) and SARS-CoV-2 across numerous continents in recent times. The difficulty with contact tracing is the quality of information received from surveyed individuals. Depending on the disease or infection, some individuals may be embarrassed or ashamed, others may lead a busy life and may not remember all the details, and others may not realise the importance of some details.

Typically, not all infections are found through contact tracing, and as R_0 increases, it is less likely that contact tracing will identify a larger proportion of the infected population.

What is contact tracing?
Contact tracing can stop an Ebola outbreak in its tracks
CDC
U.S. Department of Health and Human Services
Centers for Disease Control and Prevention
Contact tracing is finding everyone who comes in direct contact with a sick Ebola patient. Contacts are watched for signs of illness for 21 days from the last day they came in contact with the Ebola patient. If the contact develops a fever or other Ebola symptoms, they are immediately isolated, tested, provided care, and the cycle starts again—all of the new patient's contacts are found and watched for 21 days. Even one missed contact can keep the outbreak going.
EBOLA PATIENT
Isolate & Provide Care
Ask about contacts
21 days
Watch contacts for symptoms of sickness for 21 days
If contact shows symptoms isolate, test, and provide care
EBOLA PATIENT
Isolate & Provide Care
Ask about contacts
21 days
Watch contacts for symptoms of sickness for 21 days
Repeat cycle until no new patients if
21 days
contact shows no symptoms after 21 days
Contact not at risk of developing Ebola
Missed contact
May spread Ebola to new contacts
Contact tracing finds new cases quickly so they can be isolated to stop further spread.
If contact shows symptoms isolate, test, and provide care
EBOLA PATIENT
Isolate & Provide Care
Ask about contacts
If no contacts, no further spread

FIGURE 9.2.9 The CDC's contact tracing strategy for Ebola

Mass gatherings

WHO has defined a mass gathering, in the context of public health, as:

> '… any occasion, either organised or spontaneous, that attracts sufficient numbers of people to strain the planning and response resources of the community, city or nation hosting the event.'

Common mass gatherings include religious and sporting events, music concerts, festivals and political rallies (Figure 9.2.10). A major concern is that an infection acquired at a mass gathering will not only spread quickly among the attendees but be transmitted back 'home'. This could easily be within 12 hours or possibly anywhere around the world in almost 24 hours.

FIGURE 9.2.10 Mass gatherings occur for a variety of reasons, including religion, music concerts, festivals and sport.

Other than cancelling or postponing events, strategies to help manage events that involve mass gatherings include:

- political commitment
- effective planning
- surveillance systems
- documenting activities.

WHO suggests that political commitment is essential. It is highly valuable to have public health professionals involved in the highest level of organisation of events. This is an opportunity for competing agencies to work together for the benefit of the attendees of the mass gathering.

Mathematical modelling is a useful tool for planning and preparing for a mass gathering. Modelling helps organisers to predict potential communicable diseases, project outbreaks and implications for attendees and health service personnel and infrastructure. The predictions and projections can model both slow and rapid infection scenarios and consider unanticipated incidents and quality of communication between health and emergency units.

Surveillance systems can be developed to consider infections and disease as well as chemical-related events and extreme heat and cold situations. They are recommended to be adaptable to a variety of different settings.

All personnel, organisers and authorities involved in planning mass gatherings are advised to document their efforts and activities as they proceed. Records should include personal accounts of experiences and formal records including details of planning, training and exercises. Official reports and public media releases are both valuable because incident-free mass gatherings should be acknowledged as well as the problems with mass gatherings, which are often exaggerated. It is important for the organisation and operation of future mass gatherings that information and reports are evaluated appropriately.

Temperature screening

Global travel has increased considerably in recent times, which has increased the spread of disease. Many international travellers are not aware that they have an infection because they may still be in the incubation period. Other travellers may not consider themselves ill enough to not travel. They may be unaware that the strain of disease they are infected with is foreign to their destination and so they are about to expose a susceptible population. In any instance, human travellers are the greatest contributor to epidemics and pandemics.

Recent cause for concern and risk of epidemics and/or pandemics includes severe acute respiratory syndrome (SARS) (including the strain that developed into COVID–19), MERS, swine flu and bird flu (avian influenza) strains as well as other influenza strains, yellow fever and cholera. The pandemic outbreaks of SARS in 2003 and influenza in 2009 prompted many international airports quarantine stations to develop non-invasive and indirect strategies to diagnose people with infections. Infrared thermal image scanners (ITIS) became common. They attempted to use the temperature of skin to detect infected individuals, as seen in Figure 9.2.11.

FIGURE 9.2.11 Thermal monitoring attempting to detect infectious travellers

Many infections, including recent pandemics such as the SARS-CoV-2 and localised epidemics, cause the core body temperature to increase, which can be felt on the skin. One immune response to an infection is increased metabolism as the immune system synthesises antibodies and B and T cells to fight against the invasion of a pathogen. Therefore, fever is a symptom of illness and can be observed and measured by infrared thermal imaging.

The normal temperature of skin for a healthy individual is difficult to obtain. Some research has found mean skin temperatures ranging between 31°C and 37°C, while others have reported mean total skin temperature to be at 30–31°C. Skin temperature can change with environmental conditions, such as ambient temperature (e.g. in air-conditioning), humidity and wind. Also, different areas of the human body can have different skin temperatures (by up to 10–15°C); for example, skin on the foot can be a different temperature from skin on the chest.

Other factors affecting general body and skin temperature include clothing, both the number of layers as well as the type of material, the physical activity of an individual, whether they are exercising or resting, as well as whether they are carrying a load or not. The time of day is important, too, because skin temperature immediately after waking in the morning is different from 30 minutes after waking, at mealtimes or when digesting food and during an illness.

CASE STUDY 9.2.5

Infrared thermal image scanners at international airports

Infrared thermal detection systems are able to detect whether an individual has a fever but cannot detect the cause. They are able to measure skin temperature and use this to determine internal body temperature. Non-contact infrared thermometers are used away from the patient, usually between three and 15 centimetres from the forehead or the temple.

In the past border screening has been used during breakouts of various infectious diseases including, Middle Eastern respiratory syndrome (MERS) and the H1N1 influenza. Infrared thermal technology is used in at least twenty countries worldwide, including Australia, Italy, Japan, Singapore and the USA.

Two methods of detection are used: full-body scanners, which detect skin temperatures, and handheld infrared thermometers and ear gun thermometers. During the 2014 Ebola outbreak, West African airports used the latter.

In 2009, border control at Narita International Airport, Japan, implemented screening of passengers arriving from destinations with H1N1 influenza infections. The screening included a health declaration completed by passengers and infrared **thermoscanning** in the cabin of the plane before disembarking. These strategies were implemented to detect **symptomatic** passengers with infections. A total of 129 546 passengers were screened upon entry (before leaving the cabin) and nine were detected with symptoms. One passenger had the H1N1 influenza strain. Enhanced surveillance (a contact tracing method implemented separately to thermoscanning), that included mandatory reporting of individuals with infections in the community, identified 141 individuals who had travelled internationally and were not detected by entry screening. Of these 141 people, 24 had symptoms on entry and were missed by the screening.

During the COVID–19 Pandemic, fever was listed as a common symptom, but was not present in every case. Additionally, if an individual was pre-symptomatic or asymptomatic and potentially infectious, they would not be detected by thermal screening. Fevers can also be reduced in patients by using antipyretic drugs. For COVID–19, the incubation period also varied. An average of 5.2 days is given in many studies, but variations can be up to 14 days. In this time many could be infectious but still not have a detectable fever. In 2020 at Australian airports, the effectiveness of thermal passenger screening for COVID–19 was estimated to be only 46% because of the incubation period, asymptomatic cases and sensitivity issues with equipment.

A study on non-contact thermal scanning conducted during the COVID–19 pandemic found that its efficacy depended on several factors, but was not limited to: (i) the diagnostic accuracy of the devices, (ii) the prevalence of fever in the disease infected individuals.

Review

1. Determine the success rate of both thermoscanning and enhanced surveillance per 1000 passengers during the H1N1 outbreak in Japan.
2. Provide two plausible reasons why thermoscanning missed 24 individuals with symptoms on entry during the H1N1 outbreak.
3. Two reasons are given to explain why thermal scanning may not be effective. If the device used was accurate and well calibrated, and the individual being tested was exhibiting a fever, what could be another cause of error in taking measurements?
4. Explain why a varied incubation period for detecting COVID–19 makes thermal scanning at airports ineffective and research what could be done to counteract this during a pandemic.

References

Australian Government National Health and Medical Research Council. 2020, National COVID–19 Health and Research Advisory Committee, Thermal Scanning.https://www.nhmrc.gov.au/sites/default/files/2023-02/NCHRAC-Advice-13-On-thermal-scanners.pdf

Perpetuini, D., Filippini, C., Cardone, D., & Merla, A. 2021, 'An Overview of Thermal Infrared Imaging-Based Screenings during Pandemic Emergencies', *International Journal of Environmental Research and Public Health*, *18*(6), 3286. https://doi.org/10.3390/ijerph18063286

Quarantine

In most cases, placing an individual or group of people or animals into isolation from the rest of the population will stop disease transmission. **Quarantine** is isolation for a period of time in an appropriate location to stop transmission of disease. The more infectious a pathogen, meaning the higher the R_0, the more important it is to quarantine the individual or group. Again, this is due to humans being the most effective transmission pathway for pathogens and disease. With so many variables involved in disease transmission, it is best to tackle the spread of disease by enacting strategies that directly address the infected. This is also true for infected animals, especially livestock, because the economic impact can severely affect all levels of industry, from farmers to trade, and producers to consumers. It is important that those providing healthcare to the quarantined do not transmit the pathogen from quarantine to the outside population.

Quarantine can be established for individuals in specially made facilities (e.g. hospital rooms or wards, or holding pens for pets or livestock) or it can be more generalised to international borders and entire countries. Most countries have guidelines and laws about what can be brought into the country. In Australia, the Department of Agriculture and Water Resources determines the restrictions for travelling to Australia from overseas, including returning residents. This enables Australia to be a quarantined zone from certain pathogens, vectors and fomites, protecting the residents and agricultural industry.

Quarantine facilities may have a variety of specialised procedures, guidelines, equipment and personnel to handle the isolation of infected individuals or groups. This includes a:

- separate room
- separate cleaning service for consumables (e.g. bedding)
- room with its own bathroom, toilet and sink
- room with its own ventilation and air filtration system separate from the rest of the hospital
- higher level of personal protective equipment for nurses and medical staff.

It may be necessary to quarantine someone in their home. This isolates infected individuals from the public and interrupts transmission. The quarantine at home may include:

- only one adult caring for the infected individual
- isolating the infected individual from other members of the household; for example, in their bedroom with the door closed
- a separate bathroom for the infected (if possible)
- isolating the infected individual's items from everyone else's. For example, don't share towels or eating utensils, or store toothbrushes in the same holder
- using disinfectant to clean items touched by the infected individual, including bedside tables and bathroom surfaces.

If isolation at home is not possible, all family members should try to keep at least one metre away from the infected person, and/or wear a facemask.

CASE STUDY 9.2.6

Australia's response to COVID–19

Australia experienced two waves of COVID–19 during the pandemic in 2020. The first wave occurred between February and July 2020 and was mainly caused by transmission from international arrivals. The second wave occurred between July and November 2020 and was attributed to a breach of hotel quarantine in Melbourne. By November 2020, Australia had eliminated community transmission, which was then maintained during the early months of 2021 with the use of snap lockdowns.

Australia has a population of 25 million people. Over the two waves, a total of 28 000 cases of COVID–19 were recorded and 900 deaths. This is a rate of 110 cases and 4 deaths per 100 000 people.

Australia used widespread testing, contact tracing, stringent border closures, and well-timed lockdowns as key tools to effectively reduce community transmission. Additionally, international visitors were barred from entering Australia.

The second wave was considered much more severe than the first. In Victoria, the second wave lockdown was strict and lasted for much longer than any previous lockdowns in Australia. In Victoria, the following measures were taken:

- widespread closures of businesses, schools and childcare centres
- restricted operations of some industries, such as construction
- people were required to stay at home except for essential shopping by one household member and one hour of exercise daily per person
- mandatory mask wearing indoors and outdoors.
- travel restriction of 5 km radius from home
- an 8 pm curfew
- no travel between metropolitan Melbourne and rural areas, enforced by police patrol
- border closures between Victoria and surrounding states.

Tough penalties such as fines and, in some cases, jail, were used if these measures were not adhered to. The policies adopted were introduced gradually in response to the rise or fall in case numbers and recommendations from scientists and modelling programs. Restrictions eased slowly and were removed piece by piece as numbers fell and a rolling average of less than five cases was maintained over a fourteen day period. During the lockdowns, this seven day rolling average was used to monitor the rise and fall of COVID–19 cases and to keep the public informed on the impact of the restrictive COVID–19 measures.

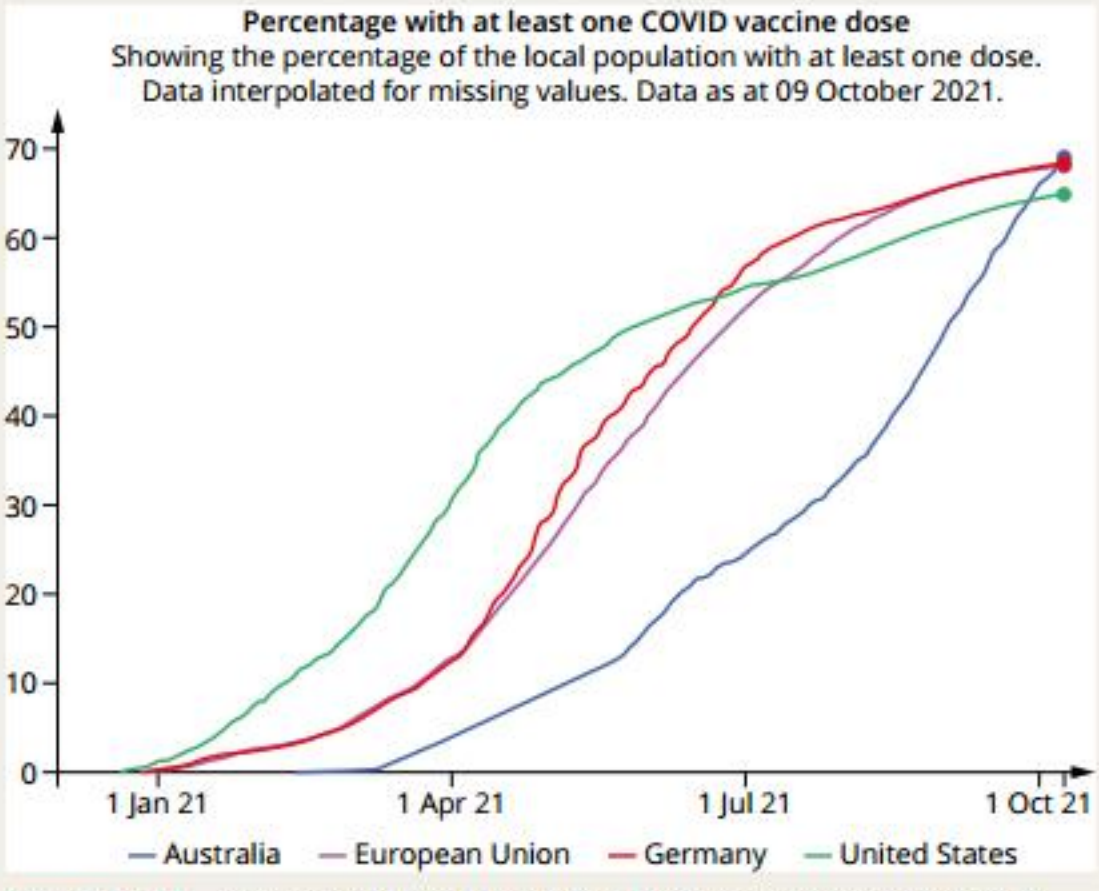


FIGURE 9.2.12 Percentage of individuals with at least one COVID vaccine dose for countries within the OECD (Organisation for Economic Cooperation and Development) between the start of the vaccine rollout and October 2021.

The vaccine program, which began in February 2021, was slow to be implemented and ill-managed. Five months into the program, only 10% of the population was vaccinated. Vaccinations were administered by doctors at health clinics. Mass vaccinations were not initially planned. The process was phased in with prioritsation of frontline workers and vulnerable individuals. The slow uptake of immunisation in the first five months is attributed to a shortage of vaccines and the AstraZeneca vaccine only being administered to over sixty-year-olds (unless individuals were granted an exception from their General Practitioner) due to the risk of blood clotting.

By July 2021, the vaccine rollout in Australia was much further behind other countries in the OECD (Organisation for Economic Co-operation and Development). Figure 9.2.12 shows the percentage of the population that had received at least one dose of a COVID–19 vaccine between its rollout and October 2021.

COVID–19 numbers increased sharply after 2021 as lockdown restrictions eased and people began socialising and returning to a pre-COVID lifestyle. By 2023, the number of cases had begun to drop and in 2024 the numbers have remained low leading up to the winter season. However, the low case numbers in 2023 and 2024 also indicate a decrease in reporting of COVID–19 cases.

Review

1. The second wave of COVID–19 was attributed to a quarantine failure in Melbourne. Research and describe the failures within the quarantine facilities that were possible causes of the second wave of COVID–19.
2. The data in Table 9.2.5 indicates that cases of COVID–19 were kept low during lockdowns. The last snap lockdown occurred during 2021. A significant rise in cases occurred in 2022 and 2023. Compare the practices used to control the spread of COVID–19 in 2020–21 and 2022–24.
3. What considerations would the Australian Government have taken into account to ensure an efficient rollout of vaccines for COVID–19?
4. In Chapter 7 you learnt about active and herd immunity. Consider Table 9.2.5 which shows the number of COVID–19 cases between 2020 and 2024.
   a. Explain how a decrease in the number of COVID–19 cases could correlate with an increase in active immunity in the community.
   b. Determine whether it can be concluded with certainty, that herd immunity to COVID–19 was reached from the data in Table 9.2.5.

TABLE 9.2.5 The number of COVID–19 cases reported in Australia between 2020 and 2024.

	2020	2021	2022	2023	2024
COVID–19 cases	29 117	538 956	10 318 311	864 552	113 207

References

Nicholas, J. 2021, Australia still lags many other countries on vaccine rollout – but it's catching up fast. *The Guardian*. https://www.theguardian.com/news/datablog/2021/oct/11/australia-still-lags-many-other-countries-on-vaccine-rollout-but-its-catching-up-fast

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CASE STUDY 9.2.7

Agricultural biosecurity against the fruit fly

Agricultural biosecurity and quarantine guidelines at Australian borders (including air freight and shipments) is a strategy to combat **agricultural pests**. Pests mostly consist of insects, such as fruit flies, which can cause damage and rotting of crops in egg, larval or adult stages. Pests can also be vectors that cause infections of plant pathogens, resulting in disease.

About 7000 hectares of mangoes are grown in Queensland. Fruit flies are one of the major pests. The Queensland mango industry produces an average of 53 500 tonnes of fruit and $140 million per year in profit, with trays being sold for $40–$50 each. The cost of managing all pests is approximately $350–$450 per hectare.

The Department of Agriculture and Fisheries (Queensland Government) provides the following recommendation for the management of **endemic** fruit flies:

> To monitor fruit fly activity, hang male lure traps under the shady canopy, where flies tend to rest. Check the number of flies trapped each week. A number of traps (one per hectare) should be hung in the middle of each large orchard block of 5.0 ha or more according to manufacturer's instructions. Control may be necessary as soon as two flies per trap per day are caught. Queensland fruit fly infests both indigenous and introduced fruits. Commercial varieties affected include abiu, apple, avocado, babaco, capsicum, carambola, casimiroa, cherry, citrus, custard apple, granadilla, grape, guava, kiwifruit, mango, nectarine, papaya, passionfruit, peach, pear, persimmon, plum, pomegranate, prune, quince, loquat, santol, sapodilla, tamarillo, tomato and wax jambu.

Scientific research was conducted to study the management of the Queensland fruit fly in Queensland in the mango industry. As the Queensland fruit fly is a major pest for many fruit and vegetable crops, it is hoped this research can inform the agricultural industry and community as a whole. The study was conducted from August 2013 to February 2014 and employed various strategies of baiting, luring and trapping flies to comprehensively evaluate the effectiveness of each.

Figure 9.2.13 shows the effect of treatments to the trunks of mango trees with fruit fly luring solutions. Bowen and Dimbulah sprayed with fruit fly lure, with Bowen spraying (treating) the trunks of every second tree in each row while Dimbulah sprayed (treated) only the perimeter trees. Mutchilba hung Cera Traps in perimeter trees. For perimeter studies, cue-lure traps were placed approximately 15 metres apart. For the study of cue-lure traps throughout a crop, there were 5–10 traps per hectare. All traps were hung within the outer canopy at approximately 1.8 metres high.

Figure 9.2.14 shows the results of specific bait height testing conducted at Cleveland. Two plant species were studied, cassava and forage sorghum, which can provide resting sites for fruit flies. Trials were performed in four 3 × 3 × 2.5 metre gauze-sided cages within a shade house at the Department of Agriculture and Fisheries Redlands Research Facility (Cleveland). In each cage, three plants were placed in each corner with only one plant type in each cage. Baits were placed at different heights in each corner. A total of 300 fruit flies, male and females, were released into each cage.

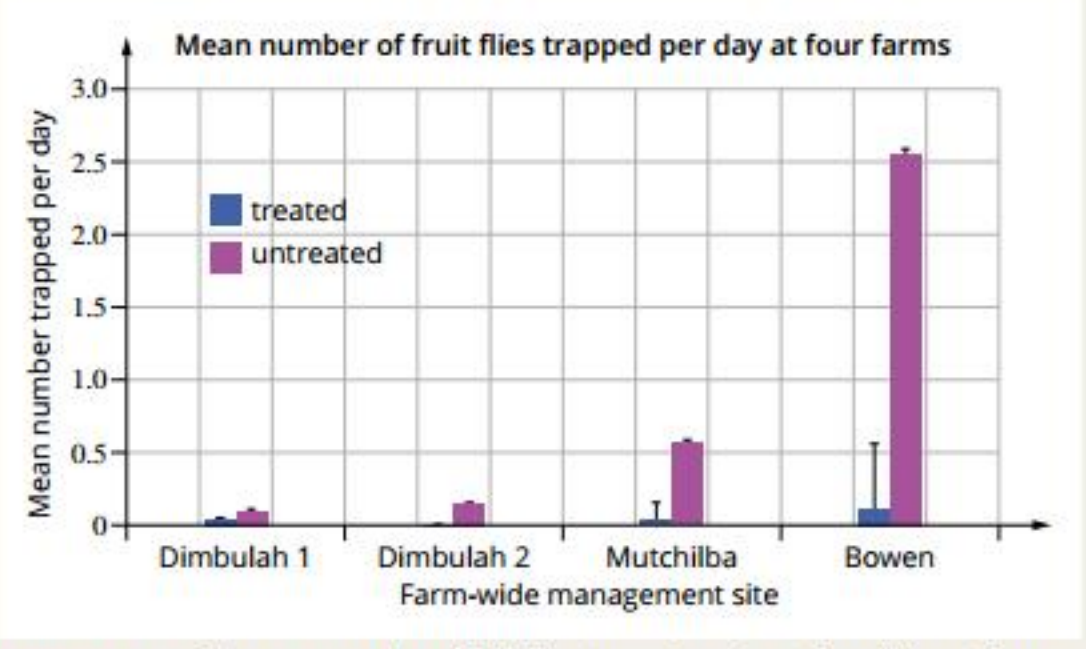


FIGURE 9.2.13 The mean number of fruit flies trapped per day at four different farms of treated and untreated crops. The Dimbulah results were affected by heavy rain, causing the spray treatment schedule to be severely altered.

Another scientific research studied fruit fly behaviour by placing a crop adjacent to forage sorghum at the Maroochy Research Facility (Nambour) (Figure 9.2.17). A row of forage sorghum was planted at one end of the crop and was 2.3 metres in height at the time of the study. Zucchini was planted in rows parallel to the sorghum in a netted area. Rows of 7, 22 and 52 metres from the sorghum were used for the trial. Both male and female fruit flies were released into the sorghum. Sample zucchini fruit were collected 1, 4, 7 and 24 hours later, and held under controlled conditions (26°C, 70% relative humidity) on drip trays in ventilated containers for a minimum of 11 days to allow eggs to develop to the pupal stage.

Review

1 Interpret the error bars for the data in Figure 9.2.13, assuming they are standard deviation.

2 Interpret the error bars for the data in Figure 9.2.14, assuming they are standard deviation.

3 Interpret the error bars for the data in Figure 9.2.15, assuming they are standard deviation.

4 Interpret the forage sorghum results in Figure 9.2.14 assuming the student *t*-test was applied, using the 2-metre-high baits as the independent variable in the null hypothesis.

5 Interpret the cassava results in Figure 9.2.14, assuming the student *t*-test was applied, using the 2-metre-high baits as the independent variable in the null hypothesis.

6 From the data presented in Figures 9.2.13–9.2.15, suggest a model for farmers to use that would minimise disease and rotting for mangoes from fruit flies.

7 Calculate the cost of managing pests at a higher level of control for the model developed for Question 6, if the typical cost of managing pests is $350–$450 per hectare.

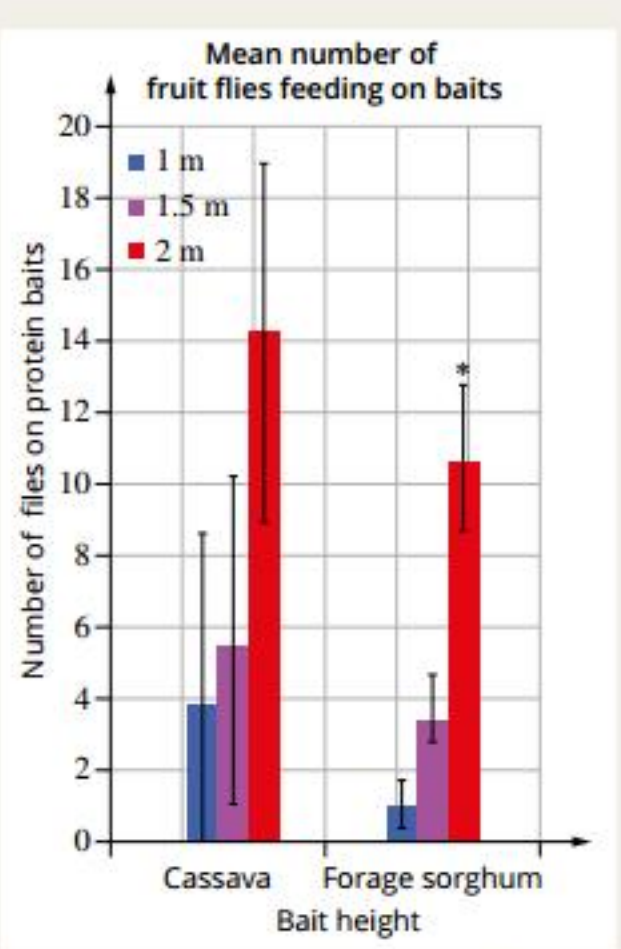


FIGURE 9.2.14 Mean number of fruit flies feeding on baits per 30 min at different heights (1 m, 1.5 m and 2 m) on cassava and forage sorghum plants. * is a statistically significant result.

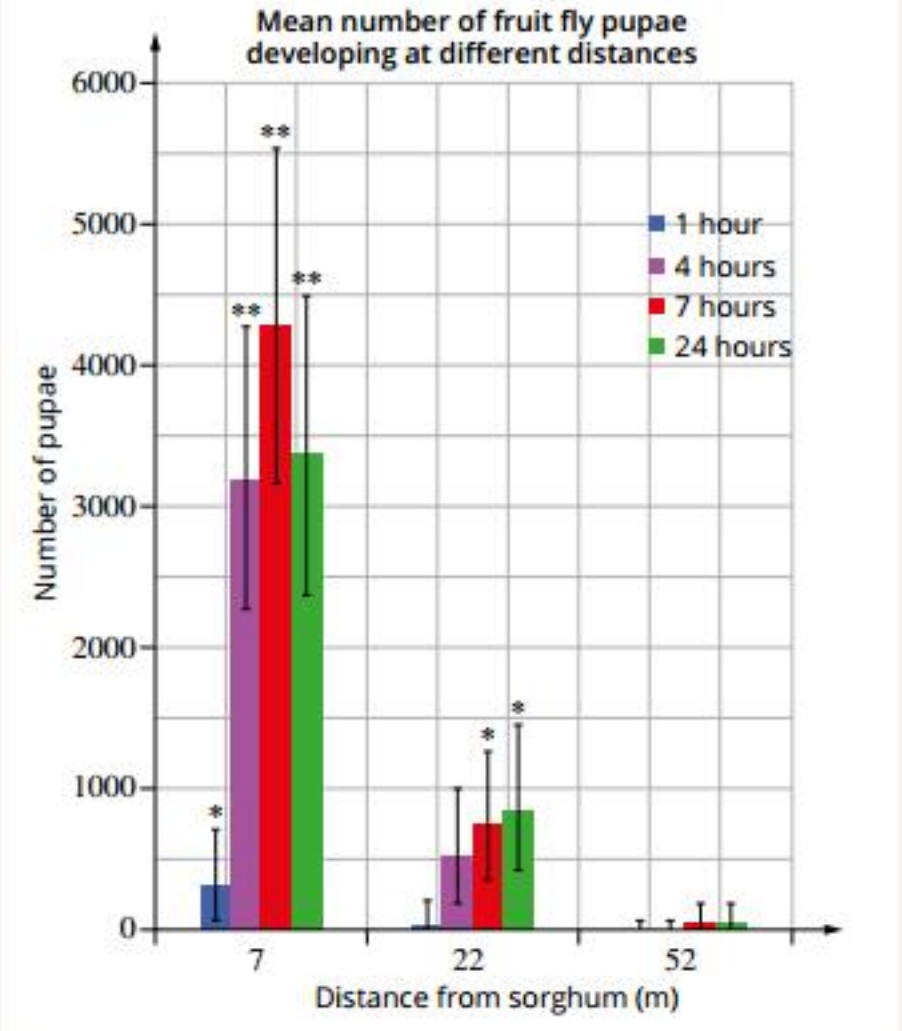


FIGURE 9.2.15 Mean number of fruit fly pupae developing in zucchinis at different distances from sorghum. The closer the zucchinis to sorghum, the greater number of pupae developing. ** is a statistically significant difference from both other distances, * is statistically significantly different from 52 m.

The costs involved in managing fruit fly pests and eradicating exotic fruit fly incursions (the importation of foreign fruit flies found in Australia) have been estimated and reported in the Australian Bureau of Agricultural and Resource Economics and Sciences report *Benefit–Cost Analysis of the National Fruit Fly Strategy Action Plan*. The cost to eradicate an **exotic incursion** fruit fly is $13.3 million per year across Australia for each exotic incursion. Table 9.2.6 shows the distribution of this cost across Australia, excluding Queensland, though the total of $13.3 million includes Queensland.

TABLE 9.2.6 Cost of eradicating an exotic fruit fly incursion into Australia, across all states excluding Queensland

State	Average annual cost ($ millions)
Western Australia	0
South Australia	1.7
Victoria	5.5
New South Wales	1.4
Tasmania	0.4
Total	8.64

Source: Plant Health Australia 2009, updated with data from state governments, where available

The report estimated that production costs of fruit and vegetables grown in fruit fly endemic areas (Queensland, New South Wales and Victoria) was $2.6 billion with 0.5–3% damaged by fruit flies. The production costs include pest management. It also reported that the value of fruit and vegetables grown in fruit fly endemic areas Australia wide is $5.3 billion, shown in Figure 9.2.16.

8 If Australia's quarantine strategy of border biosecurity did not stop an exotic Asian fruit fly incursion into Queensland, estimate the total cost over 10 years and report per year. Assume:
- eradication strategies were undertaken for 10 years without success
- the exotic fruit fly becomes a common pest like the Queensland fruit fly in 10 years, with consistent growth over the 10 years.

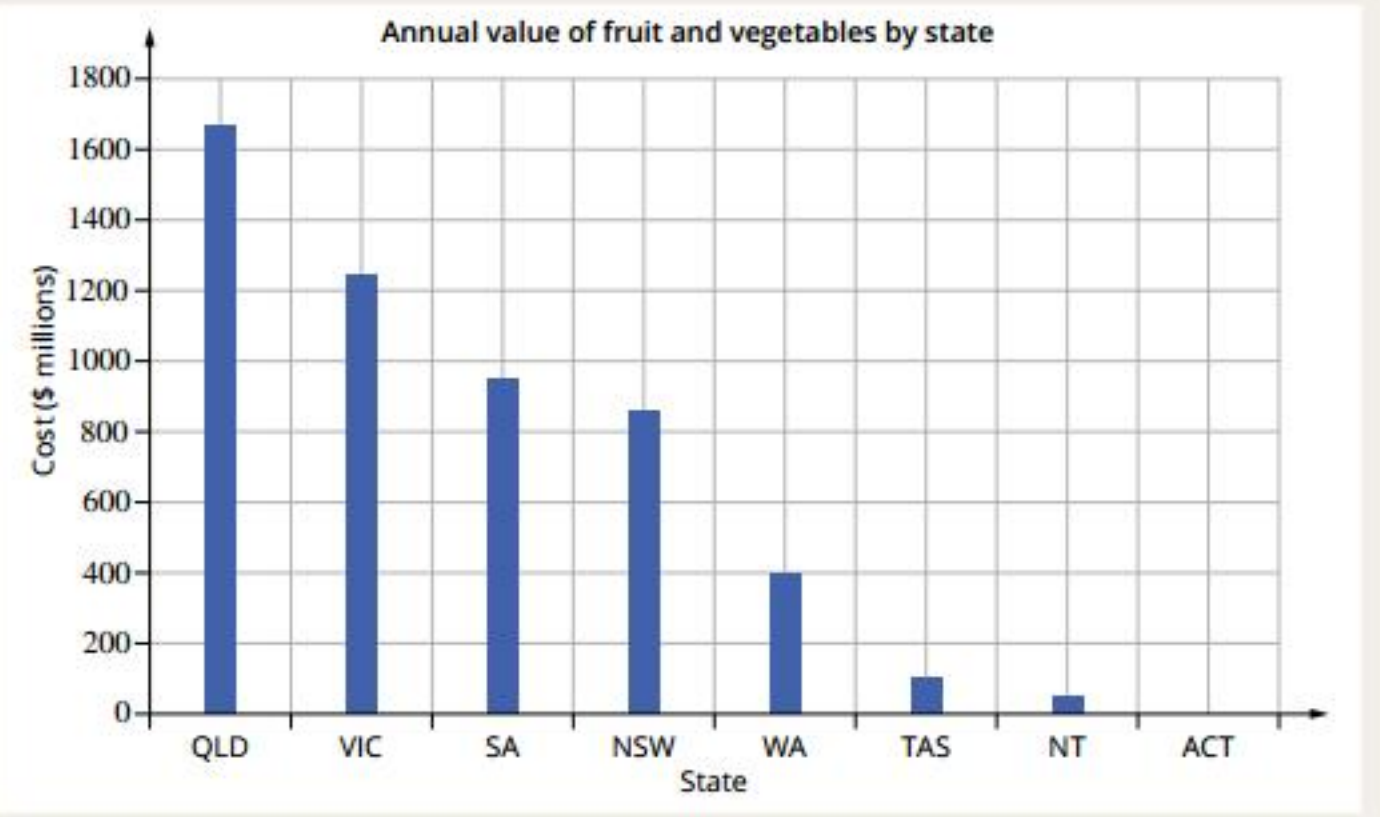


FIGURE 9.2.16 The annual value of fruit and vegetable produce in Australia by state

Review

SUMMARY

- Epidemiology is the study of diseases, how they are transmitted, and how they might be controlled.
- Various related variables must be considered, for example:
  - mechanism of transmission (direct versus indirect)
  - rate of transmission (R_0)
  - contact tracing
  - infrastructure
  - travel (regional and global)
  - herd immunity
  - hygiene
  - quarantine
  - mass gatherings.
- Epidemiology enables some prediction and control of the spread of disease using models (e.g. mathematical models).
- Developing models of disease transmission requires:
  - the understanding of transmission and methods of prevention
  - retrieving and comprehending knowledge (including immunity, pathogens and cellular biology)
  - analysing and interpreting evidence
  - applying knowledge to infer, conclude, predict, and also to create and develop preventative plans.
- Scientific skills applicable to the data test, student experiment and research investigation were used to complete case studies.

09

KEY TERMS

agricultural pest
antibacterial
asymptomatic
disease cluster
communicable disease
community immunity
contact tracing
contaminant
direct contact transmission
droplet
endemic
epidemiology
exotic incursion
fomite
herd immunity threshold
hygiene
hygiene etiquette
immunisation coverage
immunosuppressed
indirect contact transmission
mathematical modelling
morbidity
mucous membrane
pandemic
quarantine
reproduction number (R_0)
reservoir
sanitation
secondary infection
susceptible
symptomatic
thermoscanning
virulence

Data analysis

DATA SET 1

The information below applies to Questions 1–4.

Malaria is an infectious disease caused by a single-celled organism transmitted to humans via mosquitos. Figures 1–4, shows the incidence of, and deaths caused by malaria worldwide.

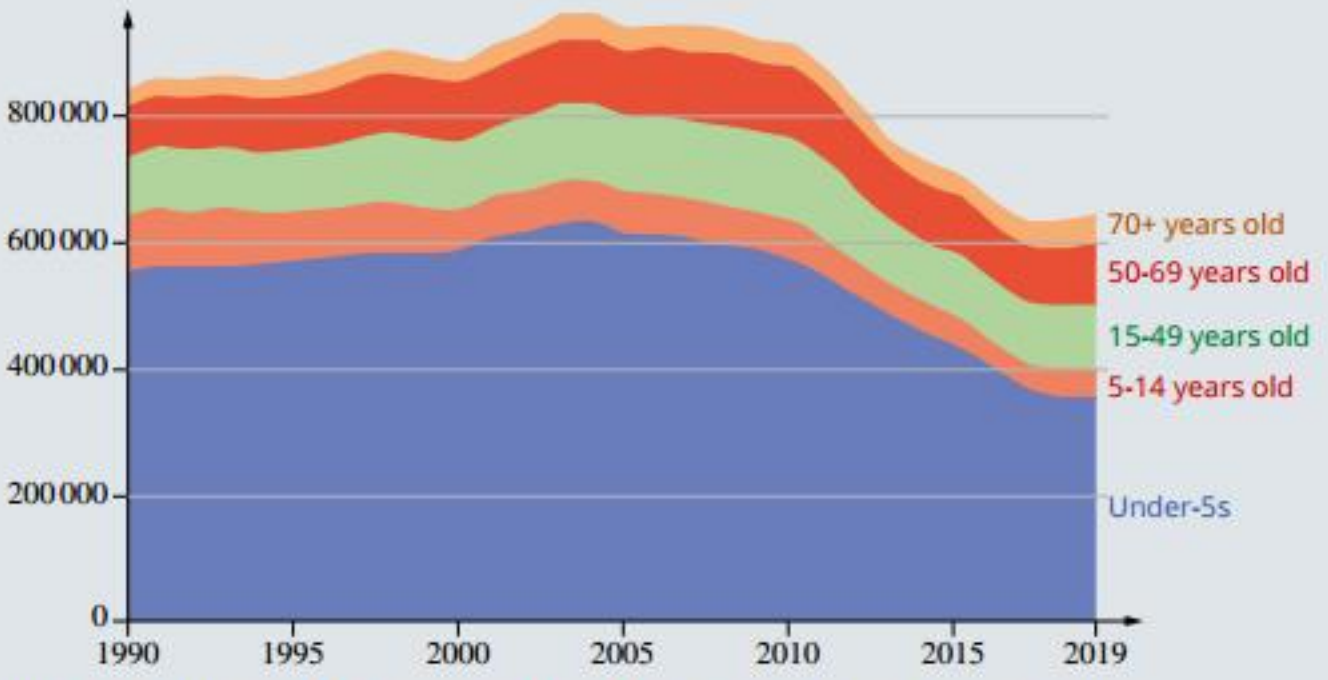


FIGURE 1 Number of deaths caused by malaria globally by age between 1990 and 2019.

Data source: IHME, Global Burden of Disease (2019)

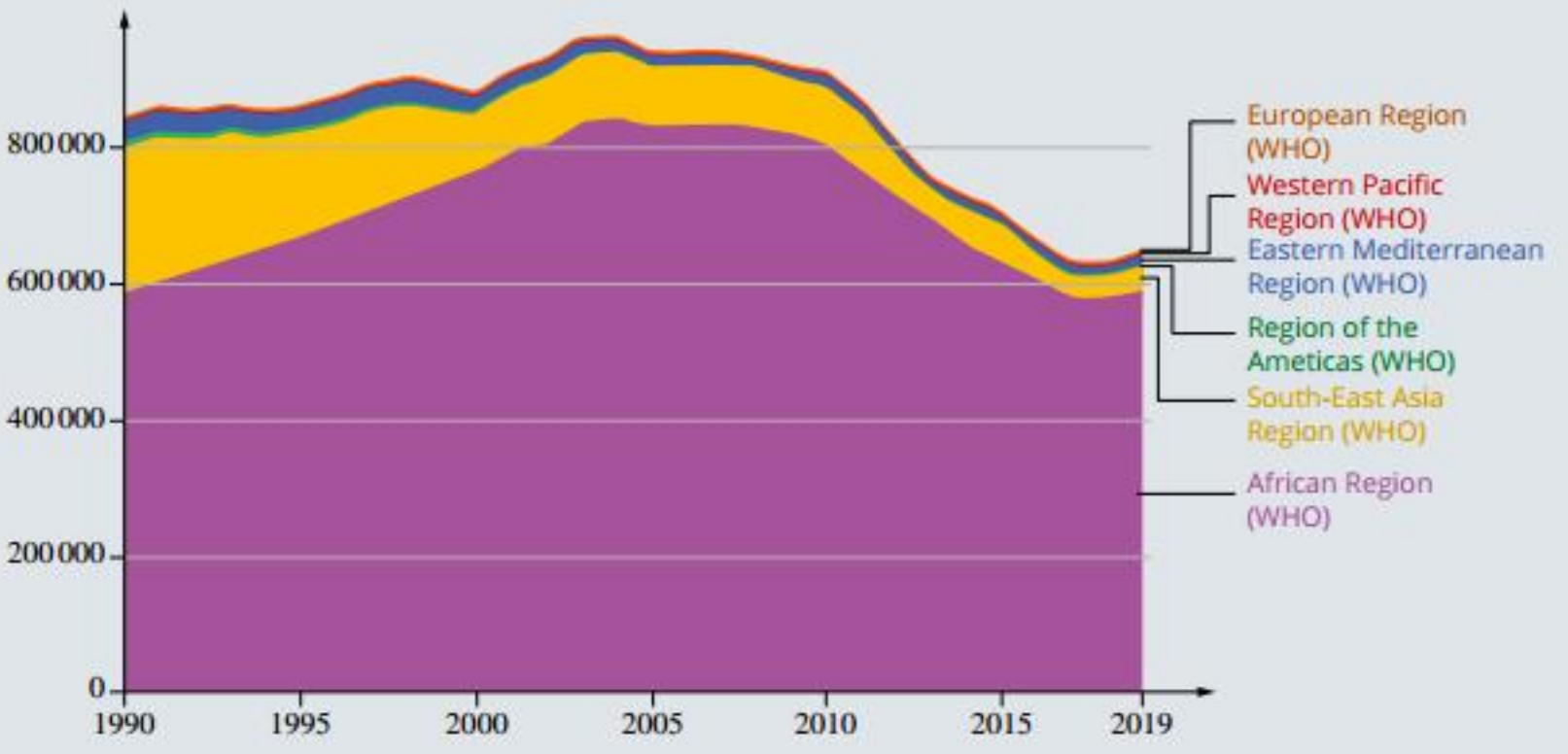


FIGURE 2 Number of deaths caused by malaria globally by region between 1990 and 2019.

Data source: IHME, Global Burden of Disease (2019)

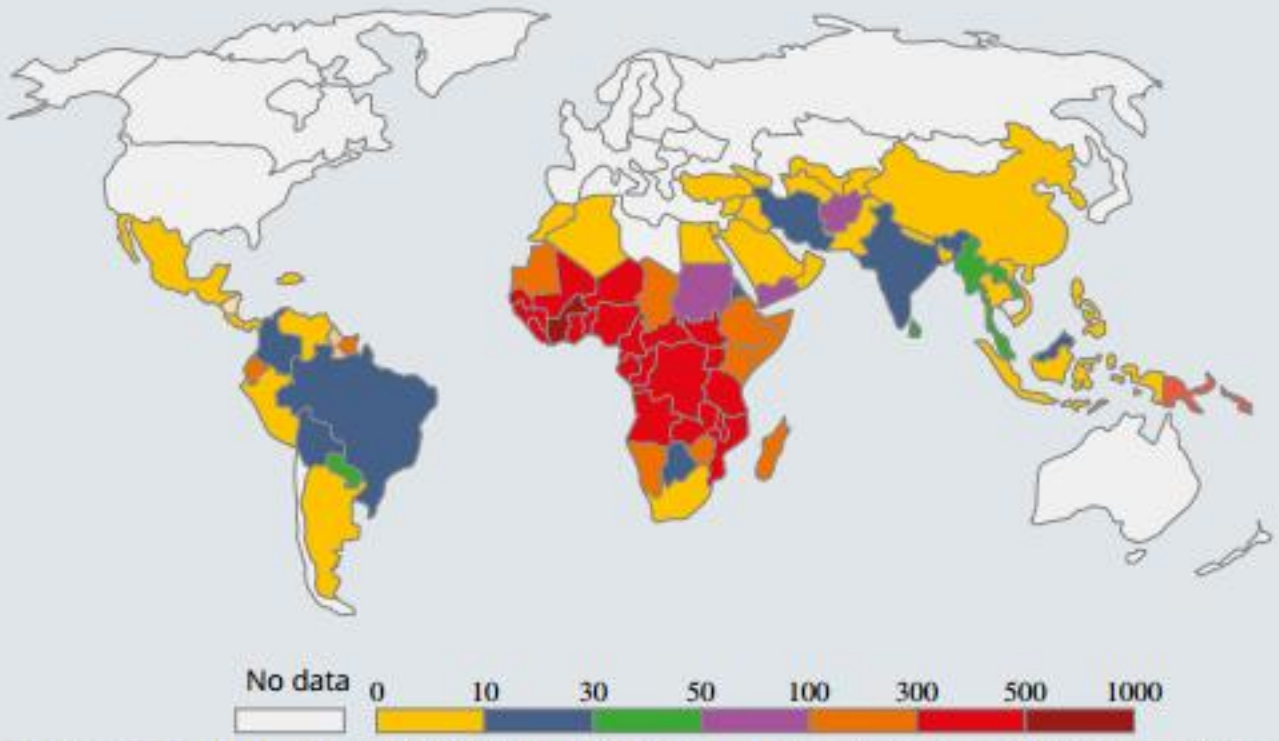


FIGURE 3 Incidence of malaria around the world in 2000. The key indicates the number of new cases of malaria per 1000 people in the year 2000.

Data source: World Health Organization (via World Bank)

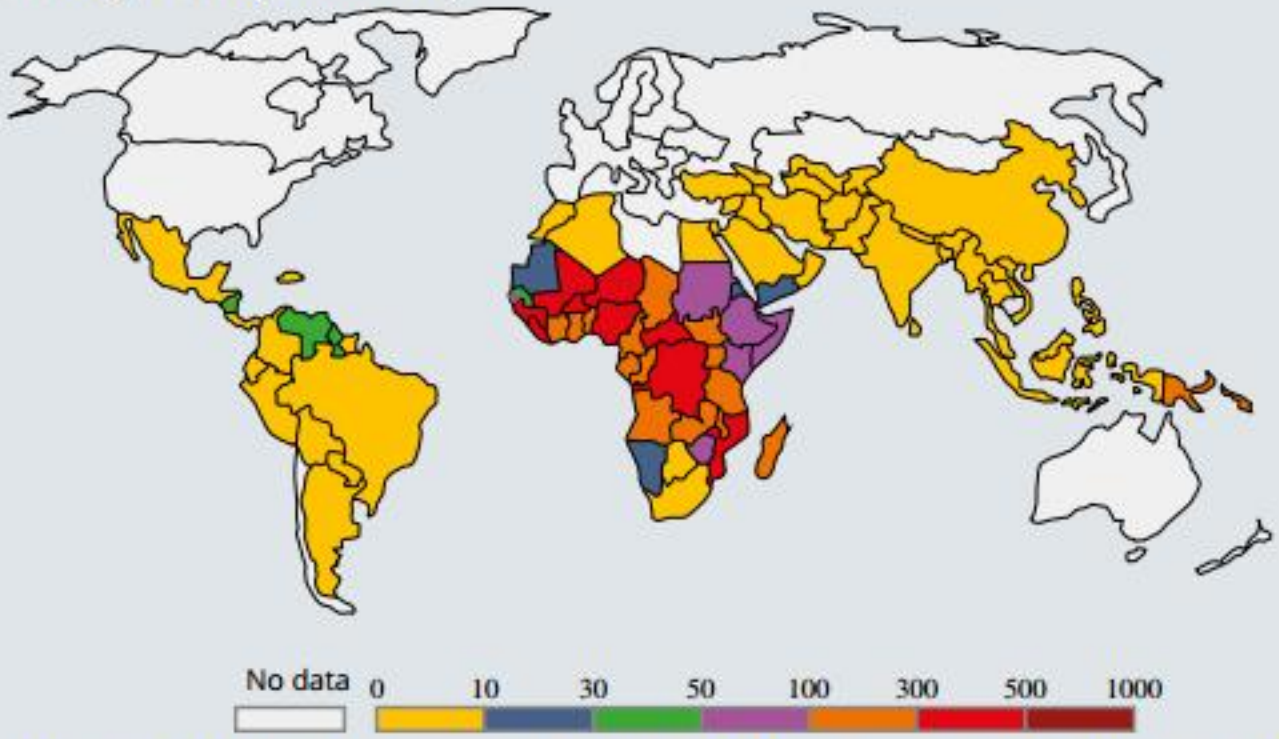


FIGURE 4 Incidence of malaria around the world in 2020. The key indicates the number of new cases of malaria per 1000 people in the year 2020.

Data source: World Health Organization (via World Bank)

Question 1 (3 marks)

Identify any trends for malaria across the world, using the data provided above.

Question 2 (3 marks)

Predict how the cases of malaria could change in the next ten years. Refer to the data in your answer.

Question 3 (2 marks)

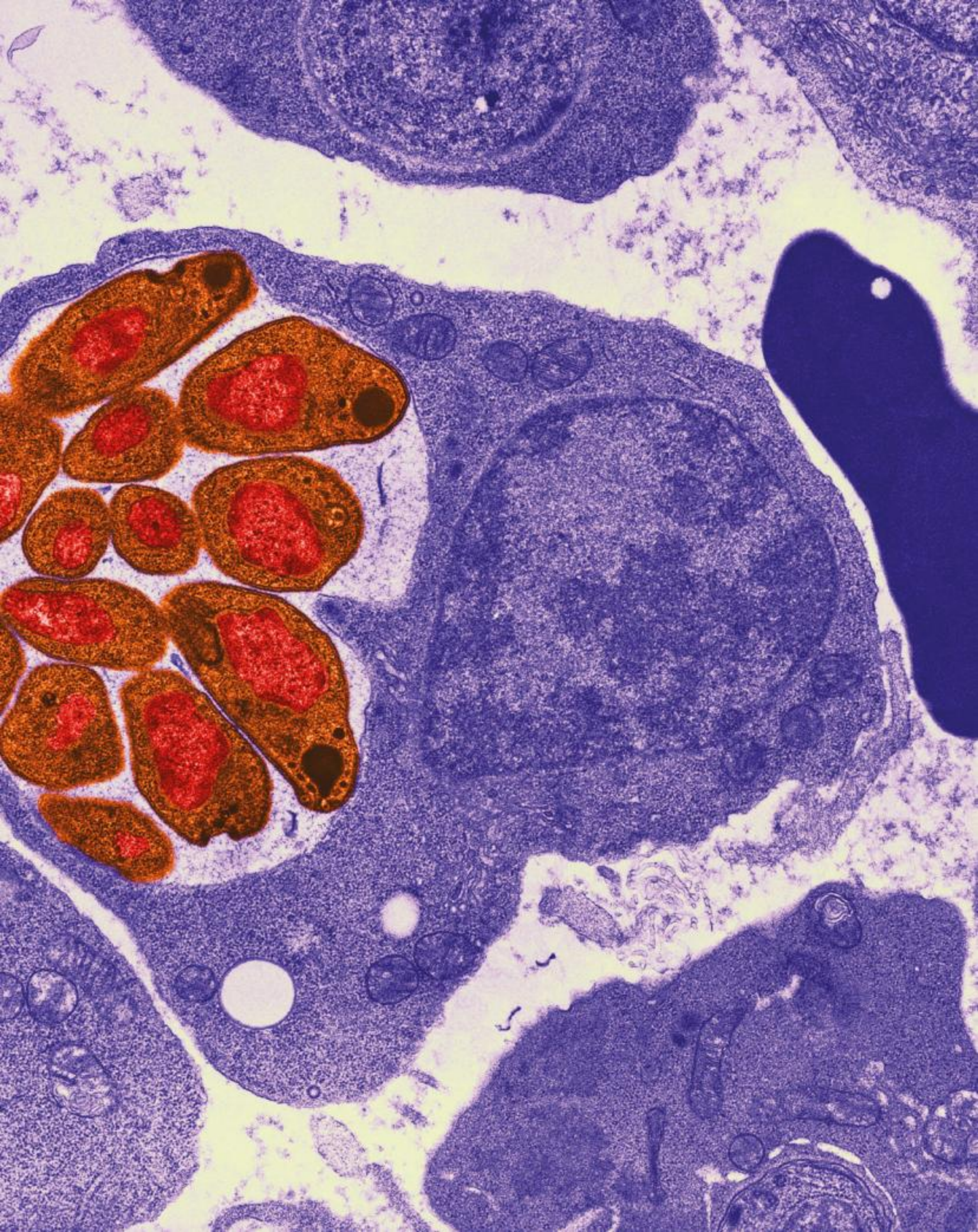
The incidences of malaria have decreased in many parts of the world. Determine why this occurred.

Question 4 (3 marks)

Children represent the largest number of deaths from malaria, particularly children under the age of five.

a Determine why this is the case.

b Identify the control measures that could be introduced to help reduce infections and death in this age group.



REVIEW QUESTIONS

Topic 1: Homeostasis—thermoregulation and osmoregulation

Multiple-choice questions

1 Select the correct statement about signalling molecules called steroid hormones.

A They are secreted by the adrenal cortex, testes and ovaries, and have their effects on target cells some distance away.

B They are hydrophilic and so cannot penetrate the cell membrane.

C They always bind to cell surface receptors to trigger chemical cascades.

D They never enter the blood of humans.

2 Identify the reason(s) most extracellular signalling molecules act on cell-surface receptors rather than intracellular receptors.

I The signalling molecules are too large to pass directly across the cell membrane.

II The signalling molecules are too hydrophilic to pass directly across the cell membrane.

III The signalling molecules are too hydrophobic to pass directly across the cell membrane.

A I only

B II only

C I and II only

D I and III only

3 Identify the best representation of the stimulus–response pathway.

A receptor → stimulus → control centre → response → effector

B stimulus → receptor → control centre → effector → response

C control centre → stimulus → receptor → effector → response

D effector → control centre → receptor → stimulus → response

4 A person is placed in water at 15°C. Assess the situation and decide how the person's body will react to maintain thermoregulation.

	Circulation to the skin surface	Activity of sweat glands	Skeletal muscle contractions
A	increases	increases	decreases
B	decreases	decreases	decreases
C	increases	increases	increases
D	decreases	decreases	increases

5 An athlete who has just completed a 20 km run will have lost an excessive volume of water through sweating. Demonstrate your understanding of water and salt regulation by selecting the statement that most accurately describes what will happen.

A Dilute urine will be produced.

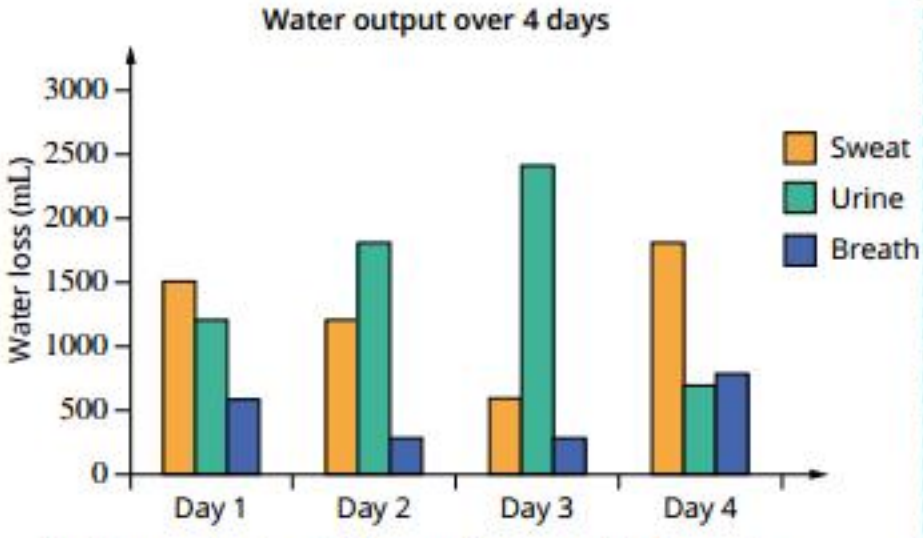
B An increased volume of urine will be produced.

C ADH will be released into the bloodstream, increasing the reabsorption of water from the collecting ducts in the kidney.

D Vasopressin will be released into the bloodstream, decreasing the reabsorption of water from the collecting ducts in the kidney.

6 A scientist was studying how water balance and environment interacted in maintaining a stable core temperature. The scientist hypothesised that environmental temperature and humidity would cause variations in how water was lost from the body.

A participant was provided with the same amount of food and drinks for four days and their water output was measured. The results of the experiment were graphed as shown below.



Environmental conditions varied over the four days. Analyse the data and select the situation that would disprove the scientist's hypothesis.

A Day 4 was hot and dry.

B Day 2 was cold and wet.

C Day 1 was cool and dry.

D Day 3 was cool and humid.

7 State where neurotransmitters are released into the synapse.

A cell body

B Golgi apparatus

C presynaptic membrane

D postsynaptic membrane

8 Salmon lay their eggs in freshwater streams, where they are fertilised. Soon after hatching, the young salmon (smolts) swim downstream and out to sea. Once in the ocean, the smolts grow into fully developed adult salmon. These adults then make their way back to their original freshwater stream to lay eggs.

Identify the best description of what would happen when the smolts move from a freshwater to a marine environment.

A The concentration of water in the blood plasma would decrease.

B A smaller volume of more concentrated urine would be produced.

C A larger volume of more dilute urine would be produced.

D Less solutes, including urea, would be filtered out of the blood.

9 After exercising in extremely hot conditions, a decrease in the concentration of sodium ions in the blood will occur. Identify which of the following will return the sodium ion concentration to its normal level.

	Aldosterone secretion	Reabsorption of sodium ions	Blood pressure
A	increased	increased	increased
B	decreased	increased	increased
C	increased	decreased	decreased
D	decreased	decreased	increased

10 Select the alternative that best depicts the correct feedback process in an endotherm.

A increase in temperature → thermoreceptor → vasodilation → hypothalamus → heat loss

B decrease in temperature → hypothalamus → vasodilation → heat gain → thermoreceptor

C increase in temperature → hypothalamus → thermoreceptor → vasodilation → hypothalamus → heat loss

D increase in temperature → thermoreceptor → hypothalamus → vasodilation → heat loss

11 After emerging from hibernation, some male red-sided garter snakes (*Thamnophis sirtalis infernalis*) can produce pheromones that mimic those produced by female snakes. This attracts other males to attempt to mate with them and, in doing so, transfer heat to them.

Identify the term given to this mode of heat transfer.

A aestivation

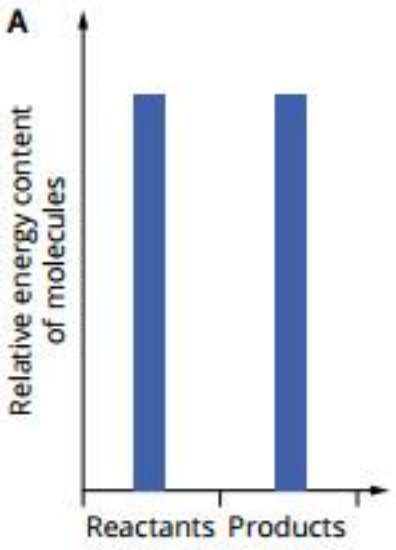
B thermogenesis

C kleptothermy

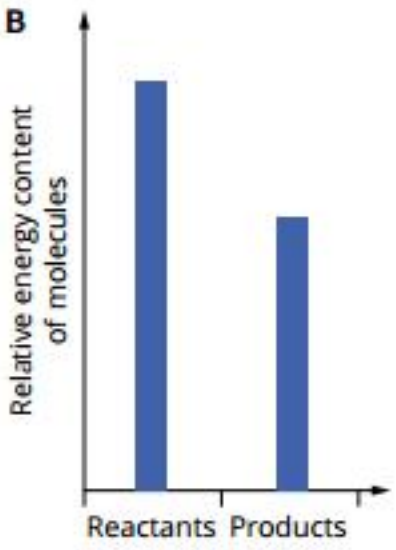
D endothermy

12 Select the graph that best represents a catabolic chemical reaction.

A



B



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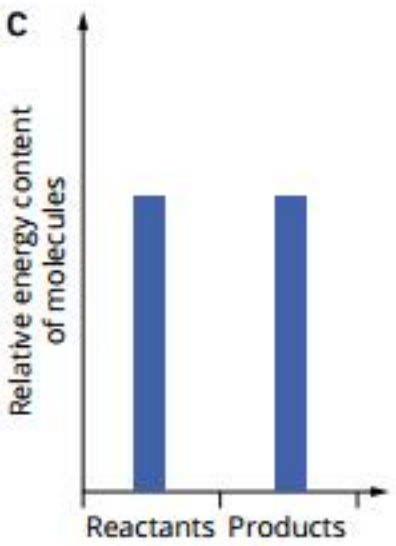

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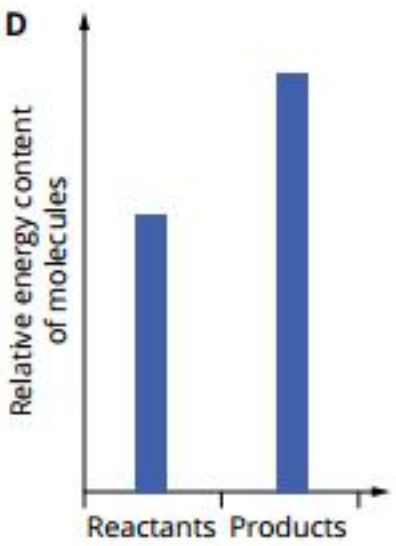


Short-answer questions

13 Define:

a metabolism

b osmoconformer

c vasodilation

d osmoregulator

e aestivation

f signal transduction

14 Explain how thermoregulation can be controlled by metabolic rate and production of TRH (thyrotropin-releasing hormone).

15 Plants produce signalling molecules called phytohormones. Auxins are a group of phytohormones and are responsible for phototropism.

a Explain the term 'phototropism'.

b i Name two sites of auxin production.

ii Describe how auxins travel to the site where they are active.

c One mechanism by which auxin causes its action is shown in the flow diagram. In the case of phototropism, the kinases at step 7 activate enzymes that soften the cell wall, making it more flexible.

Identify the three stages of signal transduction in the action of auxin.

Step 1
Auxin joins with a receptor on the plant cell membrane.

Step 2
An enzyme attached to the cytoplasmic side of the receptor is activated.

Step 3
The enzyme catalyses the change of a lipid, PIP_2, in the membrane of the cell into chemicals called IP_3 and DAG.

Step 4
IP_3 then moves to the vacuole of the cell where it joins to a receptor in the vacuole membrane.

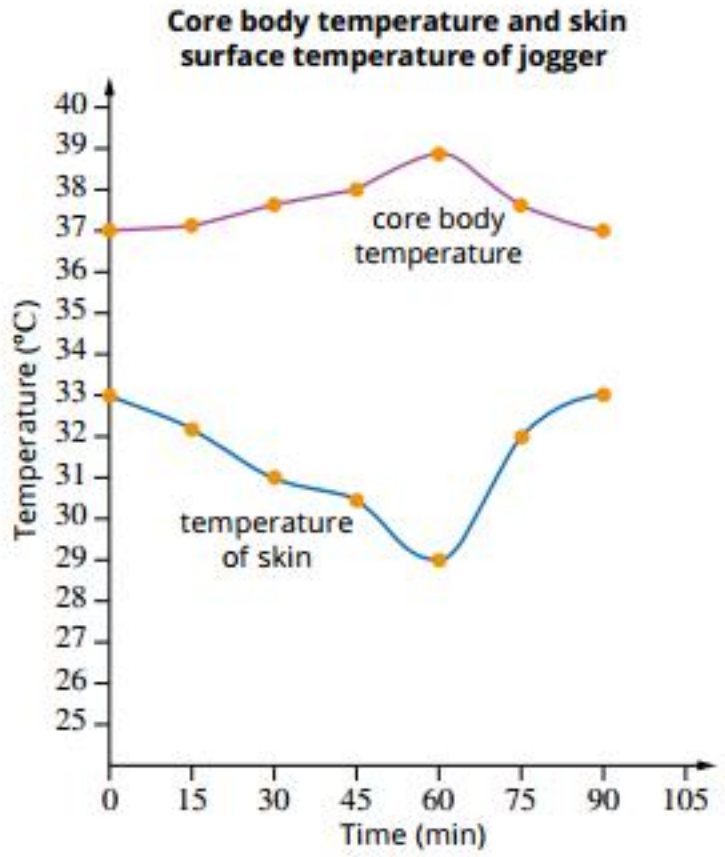
Step 5
The recognition of IP_3 by the vacuolar receptor triggers the pumping of Ca^{2+} ions out of the vacuole into the cytosol.

Step 6
The presence of Ca^{2+} ions in the cytosol activates enzymes called kinases.

Step 7
The kinases result in changes in cell activity.

d Identify the stimulus and the response of the plant at the:
- **i** cellular level
- **ii** whole organism level.

16 The diagram shows the changes in the core body temperature and skin surface temperature of a jogger. He jogged from $t = 0$ to $t = 60$ minutes. From $t = 60$ to $t = 90$ minutes, the jogger stopped and sat on a chair.

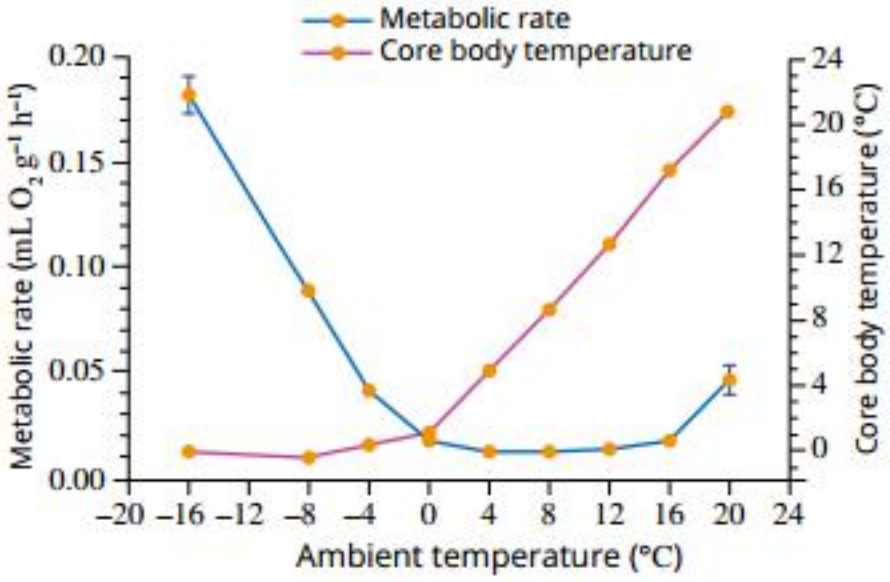


- **a** Describe the changes in core body temperature and temperature of the skin from $t = 0$ to $t = 60$ minutes.
- **b** Explain the homeostatic cause of the patterns described in part **a**.
- **c** Propose why the temperature of the jogger's skin started to rise after $t = 60$ minutes.
- **d** A phenomenon experienced by some joggers is heat stroke. Heatstroke occurs when the core body temperature rises above 40°C. Heatstroke can be fatal. Explain why heatstroke can be life-threatening, referring to enzymes in your explanation.

17 Draw a labelled diagram of an:
- **a** efferent neuron
- **b** afferent neuron.

18 **a** Acetylcholine is a neurotransmitter. Describe the role of acetylcholine.
- **b** Acetylcholinesterase is an enzyme that breaks down acetylcholine. Propose where you would expect to find this enzyme. Justify your proposition.
- **c** Redback spiders (*Latrodectus hasselti*) are found throughout Australia. They have a highly venomous bite that can result in paralysis. The situation following a bite becomes serious when the muscles of the chest are affected, causing difficulty breathing. Studies have shown that the venom causes a slow leak of acetylcholine from the presynaptic membrane of motor neurons.
  Explain how redback spider venom causes paralysis.

19 Scientists investigated the effect of ambient (surrounding) temperature on metabolic rate and core body temperature of Arctic ground squirrels (*Spermophilus parryii*). The scientists placed the squirrels in a cool environment and then slowly increased the temperature of the environment. At the same time, they measured changes to core body temperature and respiratory quotient (ratio of carbon dioxide produced to oxygen consumed) of the small mammal. Results of the experiment are shown in the following diagram.



a In order to estimate the metabolic rate of the squirrels, the scientists measured their rate of oxygen consumption. Propose why.

b Outline the effects of ambient temperature on core body temperature.

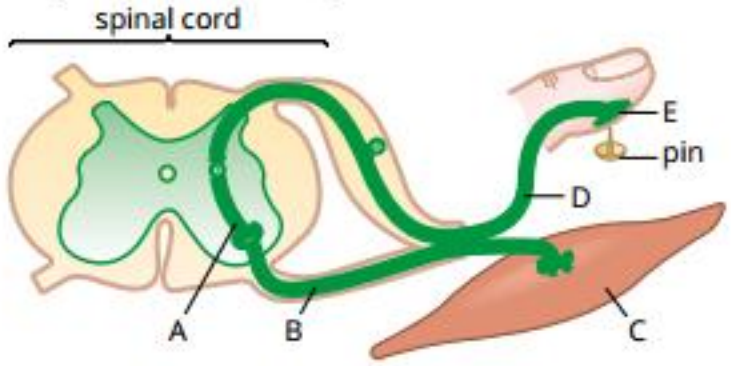
c Propose why the metabolic rate of the squirrel is high when core body temperature is low.

20 Blue whales (*Balaenoptera musculus*) are faced with a number of challenges within their fluctuating environment; one of the most challenging of which is the freezing cold water in which they live. Blue whales have developed a number of adaptations that allow them to survive in their environment.

a List some of the mechanisms that blue whales have to help regulate their body temperature.

b Briefly describe the process of countercurrent heat exchange in blue whales.

21 Carefully examine the diagram.



a Identify the structures A–E.

b Name the neural pathway shown in the diagram.

c Identify the stimulus and response shown in this situation.

d When you prick your finger with a pin, you immediately move your hand, but then you feel the pain afterwards. Explain why the two experiences are not simultaneous.

22 In a cold acclimatisation experiment, scientists noted that over a 10-day period there was increased activity in brown adipose tissue (brown fat). Explain how this could result in a reported higher level of climate comfort by the subjects of the experiment.

23 **a** The distribution of stomata across the upper and lower epidermis of four different plants was tabulated and the results are shown in the table.

Plant	Number of stomata per cm^2 on the upper epidermis	Number of stomata per cm^2 on the lower epidermis
J	0	22500
K	46000	0
L	1190	28000
M	0	0

The investigated plants were those living:

- in a forest (*Eucalyptus pilularis*)
- floating on the surface of lakes (*Nymphaea alba*)
- in deserts (*Pistacia mexicana*)
- totally submerged (*Elodea canadensis*).

Match each of the plants J–M with the environment in which it grows and explain in each case why you made your choice.

b Identify three other mechanisms that can be found in plants such as *Pistacia mexicana* that assist in maintaining water balance.

24 The use of electronic cigarettes (e-cigarettes) has increased considerably in recent years. These products have become popular, especially among the younger population and those looking for an alternative to tobacco smoking. However, concerns have been raised about their benefits to help quit smoking, as hazardous substances have been identified in e-cigarettes. Metals are one of the most concerning groups of toxins identified in e-cigarettes. The inhalation of metals has been associated with severe health effects such as respiratory disease, cardiovascular effects and the potential relationship between e-cigarette use and visual impairment. E-cigarettes may interfere with retinal vision and transmission of nerve impulses to the retina.

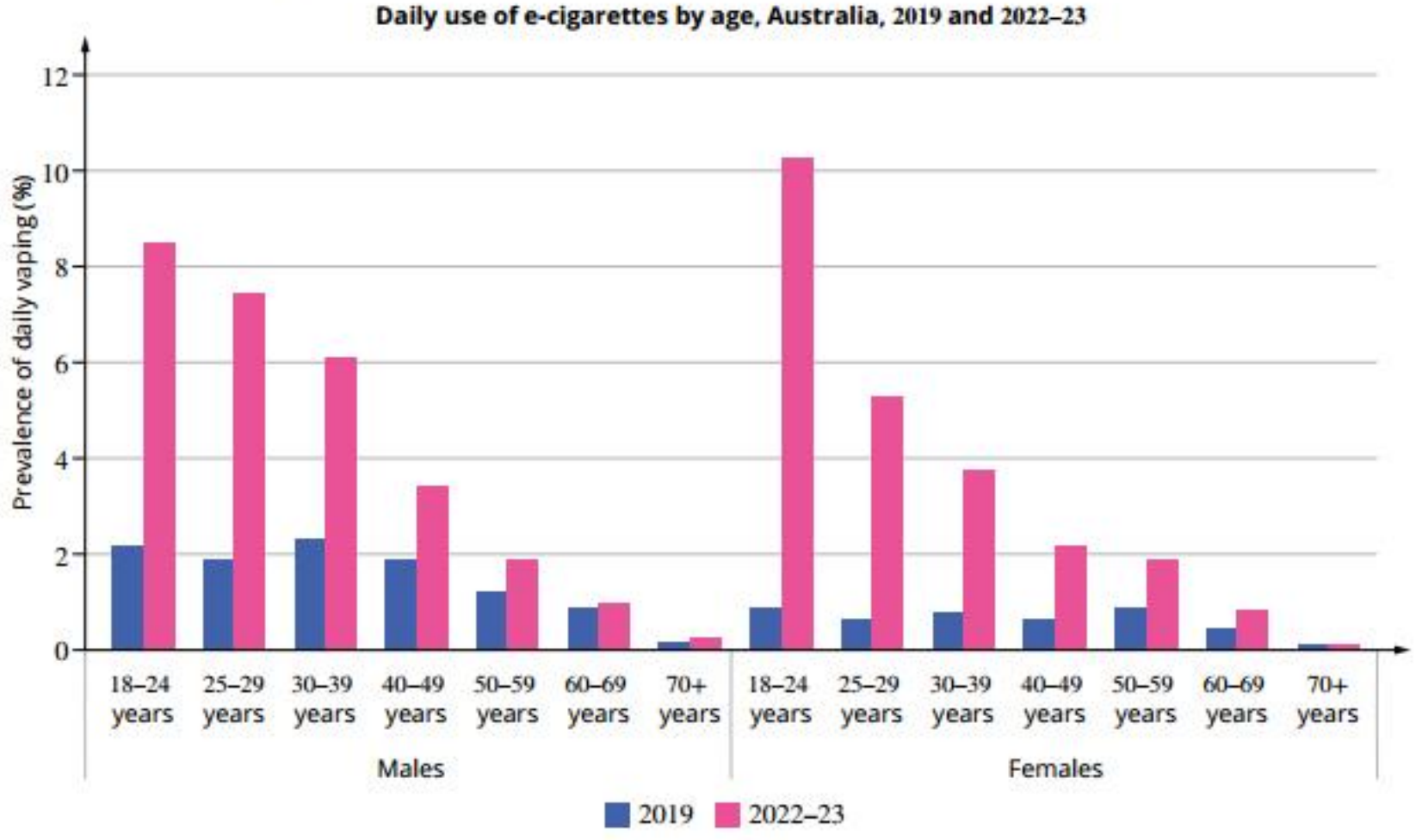
a What receptors are responsible for detecting a change in light within the retina?

b Many human regulatory systems are based on negative feedback. Explain what a negative feedback system is, and the action of the eye in response to increased exposure to light.

c How would the body's response to increased light exposure be potentially affected by the use of e-cigarettes?

d The prevalence of daily e-cigarette use for Australians in different age groups is shown in the graph below. Compare and contrast the change in prevalence of daily vaping in 2019 and 2022–23 for both male and females.

Daily use of e-cigarettes by age, Australia, 2019 and 2022–23



25 The thyroid gland produces the hormone thyroxine, which stimulates cell metabolism. It increases the rate of cellular respiration and so increases the production of both ATP and heat. The thyroid produces thyroxine when it is signalled by a hormone produced by the pituitary gland. This hormone, thyroid stimulating hormone (or TSH), is hydrophilic. The pathway to an increase in cell metabolism is shown below.

pituitary produces TSH
↓
receptors accept TSH
↓
thyroid produces and releases thyroxine
↓
body cell thyroxine receptors accept thyroxine
↓
body cells increase metabolism

a **i** Name the group of hormones to which TSH belongs.
ii State where the cellular receptors for TSH are. Explain your reasoning.

b One disease associated with the thyroid gland is Hashimoto's thyroiditis. In this disease, antibodies block the TSH receptors. Hypothesise how this affects cellular functioning throughout the body. Justify your proposition.

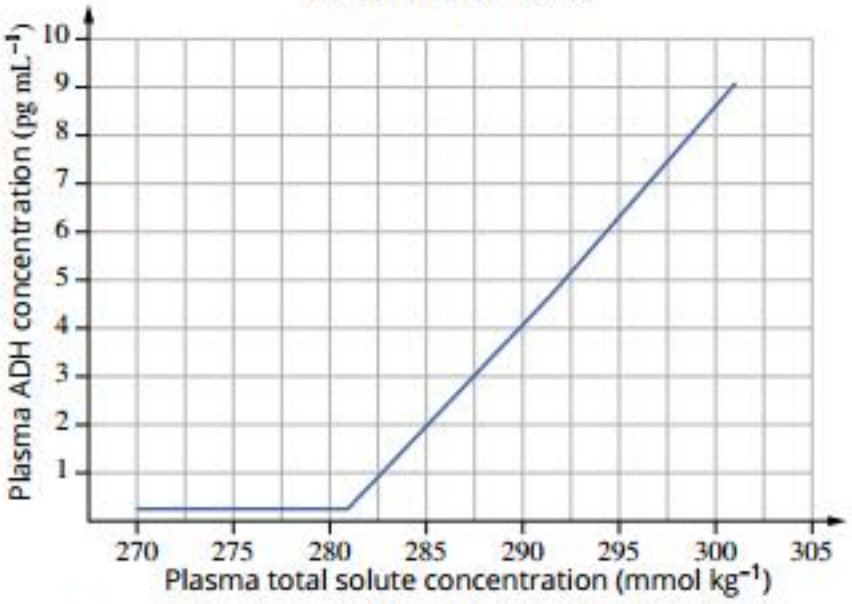
c In Graves' disease, antibodies attach to the TSH receptors and cause continuous stimulation.
i Explain how thyroid cells would respond to this.
ii Explain what effect you would expect in the body.

26 Antidiuretic hormone (ADH) is a signalling molecule produced in the hypothalamus and released by the pituitary gland.

a Name the cells in the body that respond to the presence of ADH.

b The graph shows the change in ADH as the concentration of solutes in the blood increases.

Blood ADH concentration as plasma concentration varies



i State the expected total plasma solute concentration when plasma ADH concentration is 2 pg mL^{-1}.

ii Explain how an increase in ADH concentration results in a return to a blood plasma concentration within the ideal range.

c The release of ADH is triggered when baroreceptors identify a reduction in the stretch of blood vessels and/or when osmoreceptors (which measure solute concentration in the hypothalamus) register increased concentration. Classify these two examples of receptors.

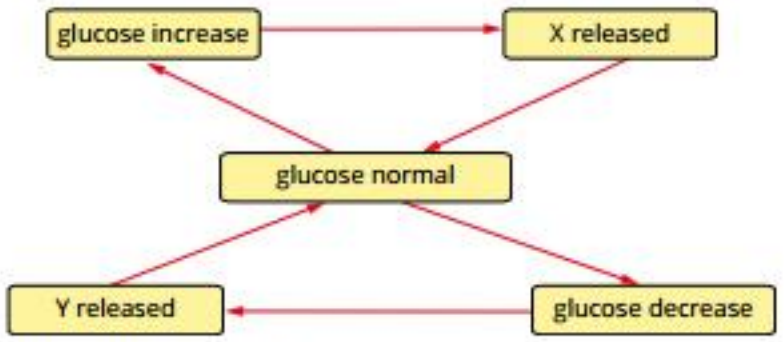
d Alcohol inhibits the production of ADH. Explain how this results in dehydration after excessive consumption.

e The spinifex hopping mouse (*Notomys alexis*) is well adapted to the desert environment where it lives. Its nephrons have very long loops of Henle. Propose how this assists the mouse to maintain water balance and live in a desert.

f When examining the concentration of ADH in the blood of hopping mice, it has been shown not to increase during periods of water deprivation. When the blood of the common house mice (*Mus musculus domesticus*) is examined under similar circumstances, they are found to have significantly higher ADH levels.

Hypothesise how hopping mice can increase their uptake of water without increasing levels of ADH while common house mice cannot.

27 The following diagram shows how the body regulates the glucose concentration in blood.



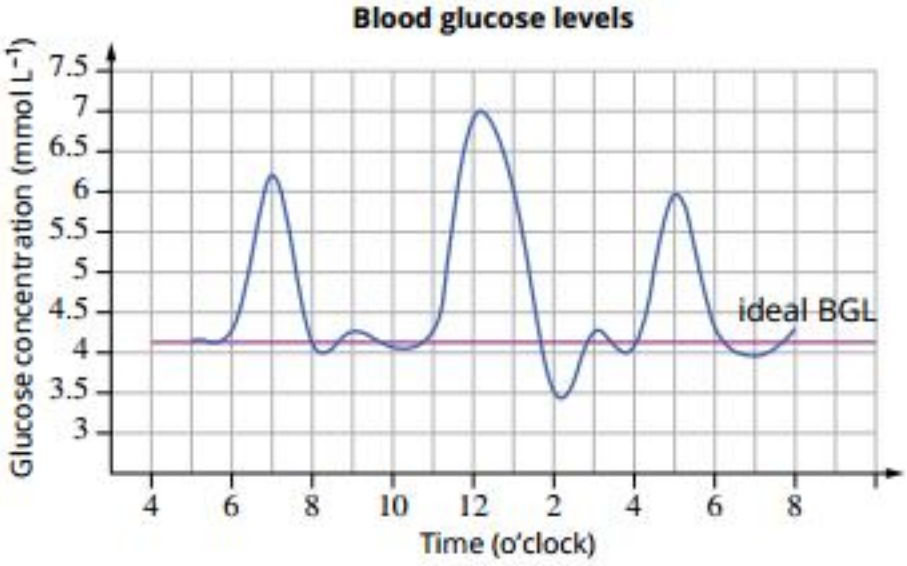
a Identify X and Y.

b i Explain how the body regulates the glucose concentration in blood.

ii Name the tissues that are the effectors in reducing blood glucose levels.

c The graph below shows the blood glucose levels (BGL) of a person throughout the day.

Blood glucose levels



i Explain at what time the person ate breakfast.

ii Explain why the BGL dips so significantly below the ideal at 2.00 pm.

28 Termites are insects that live in large social groups. In Australia, South America, and Africa, some species of termites build above-ground mounds. Because they maintain mounds at a constant temperature, these species can be subject to kleptothermy from other species. Investigate what species might be kleptotherms to termites and what these species do to obtain this energy.

Topic 2 Infectious disease and epidemiology

Multiple-choice questions

1 Viruses consist of:

A DNA surrounded by a protein coat
B RNA surrounded by a protein coat
C DNA or RNA surrounded by a protein coat
D DNA or RNA surrounded by a lipoprotein envelope.

2 Malaria is an infectious disease, with the *Anopheles* mosquito as its vector. Identify the role played by the vector.

A disease-causing organism
B blood-sucking parasite
C disease-carrying organism
D the host organism

3 Define 'pathogen'.

A A cell that destroys bacteria by injecting toxin into it.
B An organism that transfers disease from one host to another.
C An organism that can live in or on the host.
D An organism or infectious agent that causes disease in the host.

4 Identify the incorrect statement.

A Complement proteins are part of the innate immune response.
B Complement proteins attract phagocytes to the site of the infection.
C Complement proteins include antibodies.
D Complement proteins can be activated in the absence of antibody–antigen reactions.

5 Identify which of the following statements correctly states how natural active immunity is achieved.

A exposure to live or attenuated vaccines
B infection by particular bacteria or virus
C the administration of antibodies or antitoxin specific to a particular microorganism
D adequate breastfeeding in newborn infants

6 Identify the best protection for newborn babies from pertussis until week 8.

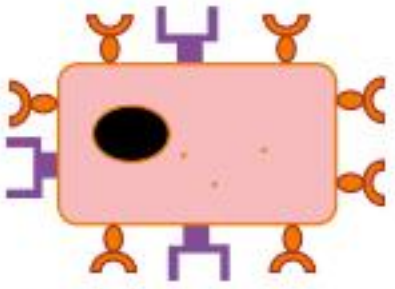
A active immunity
B herd immunity
C passive immunity
D specific immunity

7 Identify which of the following represents the correct sequence of events when the body is responding to a bacterial infection.

I antigen presentation by macrophages
II activation of B cells
III activation of helper T cells

A I, II, III
B I, III, II
C III, II, I
D II, III, I

8 The pathogen shown below enters a human body and antibodies are produced against it.



Select the antibody that would be made in response to the pathogen.

A

B

C

D

9 Identify how skin and mucous membranes act as barriers to infection.

	Skin	Mucous membranes
A	Skin is tough and forms an effective physical barrier.	Mucous membranes are thick and elastic, so pathogens are repelled.
B	Phagocytes on the skin surface trap pathogens.	Mucus is moved out of the body by the beating of hair-like cilia.
C	Skin is tough and forms an effective physical barrier.	Pathogens are trapped by sticky mucus.
D	Phagocytes on the skin surface trap pathogens.	The acidity of mucus kills harmful bacteria.

10 Select a correct explanation for why there are many different types of B lymphocytes in the body.

- **A** Each type can recognise one specific antibody and produces a specific antigen against it.
- **B** Each type can recognise one specific antigen and produces a specific antibody against it.
- **C** Each type can recognise one antigen and engulf it by phagocytosis.
- **D** Each type can recognise one antibody and engulf it by phagocytosis.

11 Plants have defences against invading pathogens. Select the alternative that is not a plant defence against pathogens.

- **A** chitinases, which disrupt the cell membranes of fungi
- **B** perforins, which rupture the cell membranes of bacteria
- **C** tannins, which are toxic to insects
- **D** defensins, which are toxic to microbes

12 Identify the correct description of why the immune system may reject an organ transplant.

- **A** The transplanted organ contains white blood cells, destroying the recipient's immune system.
- **B** Blood from the transplanted organ will cause a deterioration of the recipient's tissues.
- **C** Antibodies from the donor will react with the antibodies from the recipient.
- **D** The donor organ carries markers on the cell surface that are recognised as foreign by the recipient's T cells.

13 A baby was born severely jaundiced and anaemic. The baby recovered after a blood transfusion, but concerned doctors tested the blood of both the baby and her mother. It was discovered that the baby and her older brother were positive to the Rhesus factor, while their mother was Rhesus-negative.

Deduce the reason why the baby was severely jaundiced and anaemic.

- **A** The mother's antigens recognised the baby's antibodies as foreign and attacked them.
- **B** The mother had an adaptive immune response to the Rhesus factor in the blood of her first child, and memory cells triggered an immune response to the Rhesus factor in the blood of her second child.
- **C** The baby developed antibodies in response to her mother's Rhesus-negative blood and caused an immune response.
- **D** The Rhesus antigen present on the mother's red blood cells caused an immune response to the baby's blood, which was free of the antigen.

14 Identify which of the following types of data it would be necessary to collect for an epidemiological study.

- **A** gender
- **B** age
- **C** date of diagnosis of disease
- **D** all of the above

15 Identify which of the following methods would be ineffective at preventing the spread of an airborne contagious disease.

- **A** washing hands after using the bathroom
- **B** wearing a mask in public places and on public transport
- **C** coughing into a tissue and disposing of it
- **D** staying at home until no longer infectious

Short-answer questions

16 Match the correct definition with the following terms.

Term	Definition
Pathogen	An organism that carries a pathogen between hosts.
Infectious	A pathogen spread from a non-human animal to a human.
Epidemic	An infectious agent that causes disease.
Vector	A rapid and widespread of disease at a certain time due to a large number of hosts.
Zoonoses	A disease caused by pathogens and passed from one organism to another.

17 Identify the type of pathogen that causes the disease and list the features of that type of pathogen in order to complete the table.

Disease	Type of pathogen	Characteristics of the pathogen
Tetanus		
Malaria		
Thrush		
Leaf gall		
Trichinosis		
Mad cow disease		
Common cold		

18 Nutritional disease is common in many countries.

- **a** What is a nutritional disease?
- **b** From your definition in part a, deduce whether nutritional disease would be seen in Australia.
- **c** Explain your answer to part b.

19 How does each of the following factors contribute to the virulence of a disease caused by a pathogen?

- **a** adhesin factors
- **b** invasion factors
- **c** capsules
- **d** toxins

20 **a** Define 'inflammation'.

- **b** Outline the steps involved in initiating an inflammatory response.

21 **a** Define 'chemical barrier' and provide an example for both plants and animals.

- **b** Explain why taking a course of antibiotics can disrupt microbiological barriers in animals.

22 **a** Distinguish between complement proteins and cytokines.

- **b** Identify the type of defensive molecule that includes interferons.

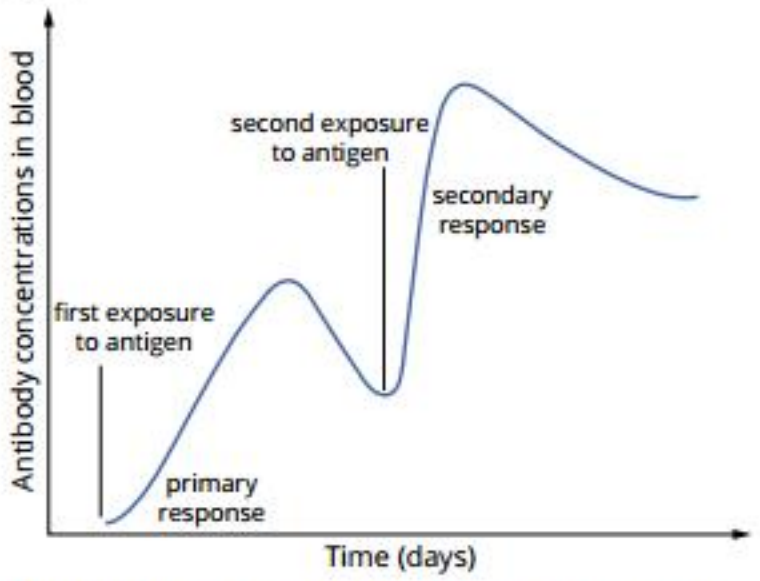
23 **a** Distinguish between bacteria and viruses.

- **b** Briefly outline why antibiotics are effective against bacteria but not viruses.

24 Explain where adherence factors are found and what their function is.

25 Most people in Australia are vaccinated against measles in childhood. The vaccination schedule requires children to be given one injection at 12 months and a second injection at 18 months.

The antibody response to the vaccinations is shown in the graph below.



- **a** Explain why the secondary response is so much greater than the primary response.
- **b** T lymphocytes play a significant role in the adaptive immune response of the body against viruses such as the one that causes measles. Describe the role of T cells in immunity.
- **c** In February 2016, Department of Health statistics showed that 93.58% of 5-year-old children in Victoria were fully immunised against measles. In some areas, the immunisation rate is as low as 73%. Explain why this statistic would be concerning to epidemiologists.

---

26 The calculation of R_0 needed to predict the spread of disease is given by the formula:

$$R_0 = r \times c \times d$$

where r is the transmissibility of the pathogen, c is the average rate of contact between susceptible and infected individuals and d is the duration of infectiousness of the disease.

Consider the information given in the table for some hypothetical emerging diseases.

- **a** Calculate the value of R_0 for each of the diseases by using the formula given.
- **b** Identify which disease(s) is diminishing in the population.
- **c** Identify which of the diseases most probably is demonstrating the effects of herd immunity. Explain your reasoning.
- **d** **i** Propose which of the diseases would be of most concern to health authorities. Justify your proposal.
  - **ii** Explain three public health initiatives which could be introduced to reduce the spread of this disease.
  - **iii** List at least one initiative that is unlikely to have much effect.

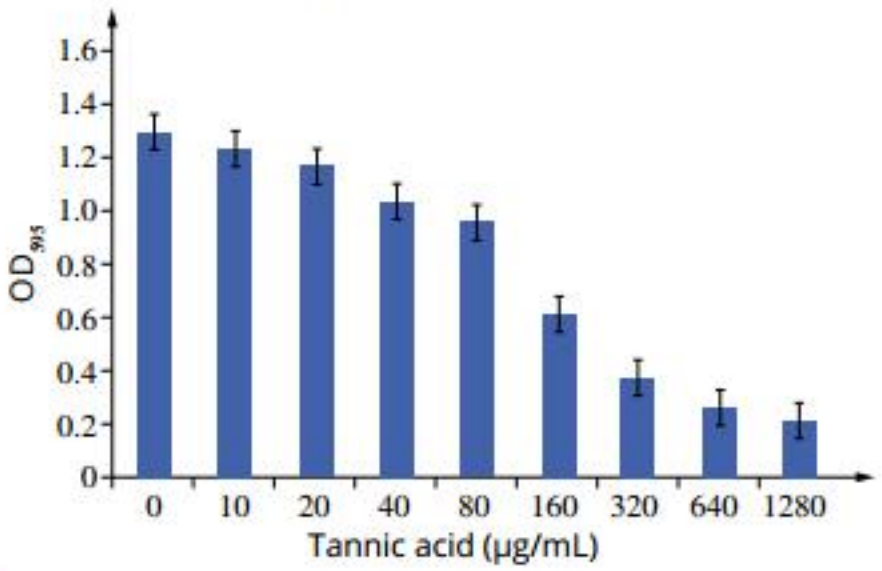
Disease	Probability of infection given contact between an infected person and a susceptible person (transmissibility)	Rate of contact (probability) between an infected and a susceptible person	Duration of infectiousness (days)	Mortality (%)	Pathway of spread	Type of pathogen
A	0.9	0.95	7	4	foodborne	bacterium
B	0.2	0.05	14	30	airborne droplets	virus
C	0.35	0.4	20	0.2	insect vector	protozoan
D	0.8	0.6	11	12	contaminated water	bacterium
E	0.1	0.9	6	0.2	direct contact	virus

27 Explain the cause and method of transmission of HIV.

28 Evaluate the ability of quarantine to prevent the entry and spread of disease into Australia, using examples to illustrate your answer.

29 Tannic acid is a chemical produced by plants which was investigated for its bactericidal properties against *Pseudomonas protegens*. The bacteria were grown in test-tubes containing growth medium and tannic acid at concentrations as indicated on the *x*-axis of the graph below. Growth of the bacteria was determined by measuring the optical density of the medium after 24 hours. Higher density indicates more bacterial growth. The results are shown in the graph.

Does the data support the hypothesis that tannic acid has bactericidal properties? Justify your answer.



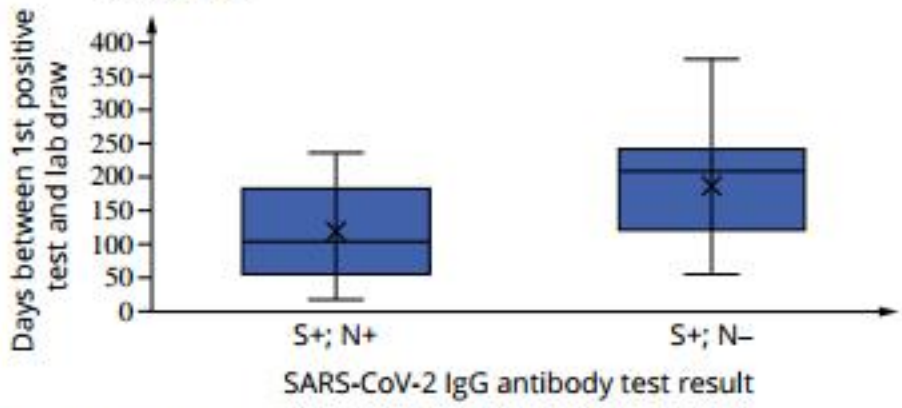
30 Many natural therapies websites describe eucalyptus oil as an excellent antiseptic.

- **a** Distinguish between antiseptics, disinfectants and antibiotics.
- **b** Design an experiment, using paper discs, to test whether the claim that eucalyptus oil is an effective antiseptic is valid.
- **c** Predict the outcome if the claim that eucalyptus oil is an effective antiseptic is valid.
- **d** Explain why it is necessary to include a paper disc without eucalyptus oil.

31 Cytokines are signalling molecules. Several different types of cytokines are used in the immune response. Discuss the role of three cytokines in the immune response, including what, if any, cells they work with and what they do.

32 From May to July 2021 a study was undertaken to determine the level of COVID–19 infections in children in Colorado, USA. One aspect of the study was the analysis of blood samples for two different antibodies, IgG against the spike protein (S) and IgG against the nucleocapsid protein (N). Samples were taken at varying times from the confirmation of infection, as shown in the box plots below.

- **a** **i** What conclusions can be drawn about antibody persistence from this data?
  - **ii** How could this influence the production of vaccines?
- **b** The mRNA vaccines currently in use cause the recipient to make spike proteins, resulting in an immune response. What difference would you expect to see in boxplots from mRNA recipients compared with the individuals tested in this study? Explain your reasoning.



33 Research anaemia and answer the following questions.

- **a** Assess how is it related to nutritional disease.
- **b** Propose who in the community might suffer from anaemia and why.
- **c** Can anaemia be linked to other chronic diseases?
- **d** Through your research, identify how anaemia may be treated.

34 Pathogens can be cellular or non-cellular.

- **a** List at least two examples of each type of pathogen.
- **b** Create a comparison table that identifies similarities and differences between these pathogens looking at the following characteristics:
  - How does this pathogen infect its host?
  - How does this pathogen affect its host?
  - How can this pathogen be managed?

35 Self-tolerance is important for a B cell to develop.

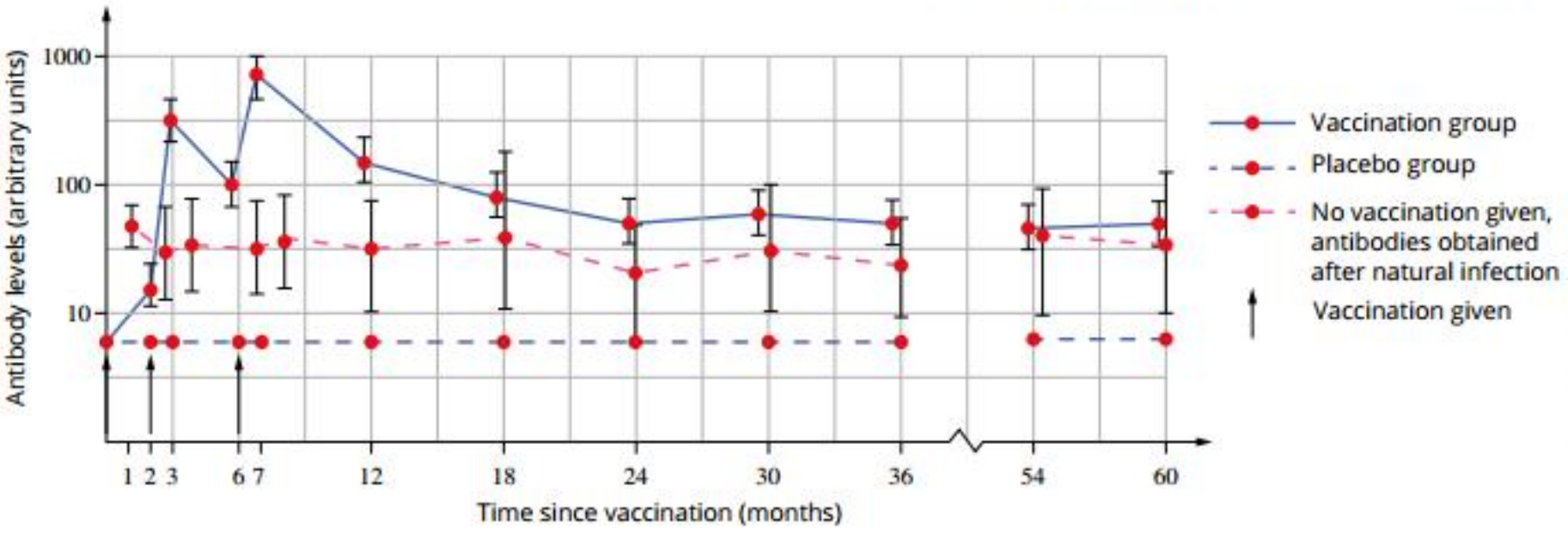
- **a** Explain what is meant by the term 'self-tolerance'.
- **b** Appraise the importance of self-tolerance in B cells.

36 The graph below shows the antibody levels for human papilloma virus (HPV)18 in women aged between 15 and 26 years. The level of antibodies in the blood was measured for three different groups of women: the first group were given the vaccination against HPV (Gardasil®), the second group were given a placebo, and the third group consisted of individuals who had obtained antibodies after natural infection.

a State when the vaccination group had the highest levels of antibodies for HPV18.

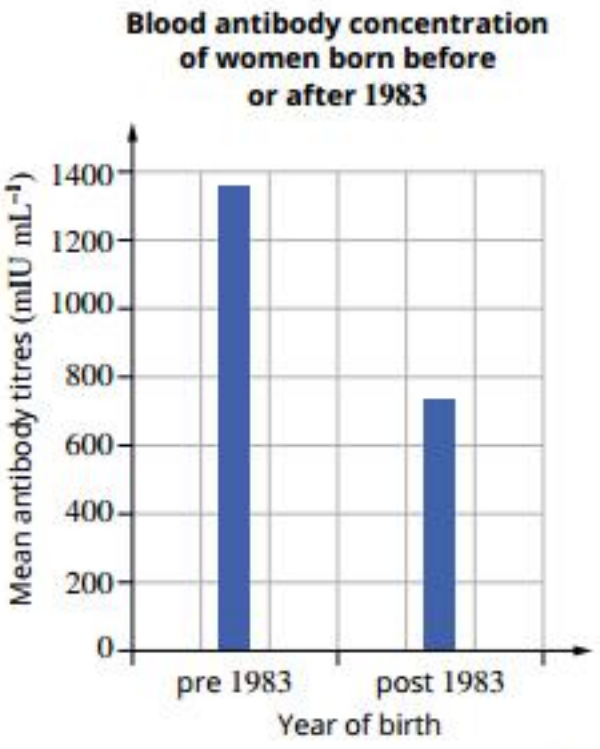
b Contrast the levels of antibodies for HPV18 between the three groups.

c HPV vaccination is often given as three injections over 6 months: the second dose is 1–2 months after the first dose, and the third dose is 6 months after the first dose. Based on the results of the study, explain why it is essential that individuals take all three doses.



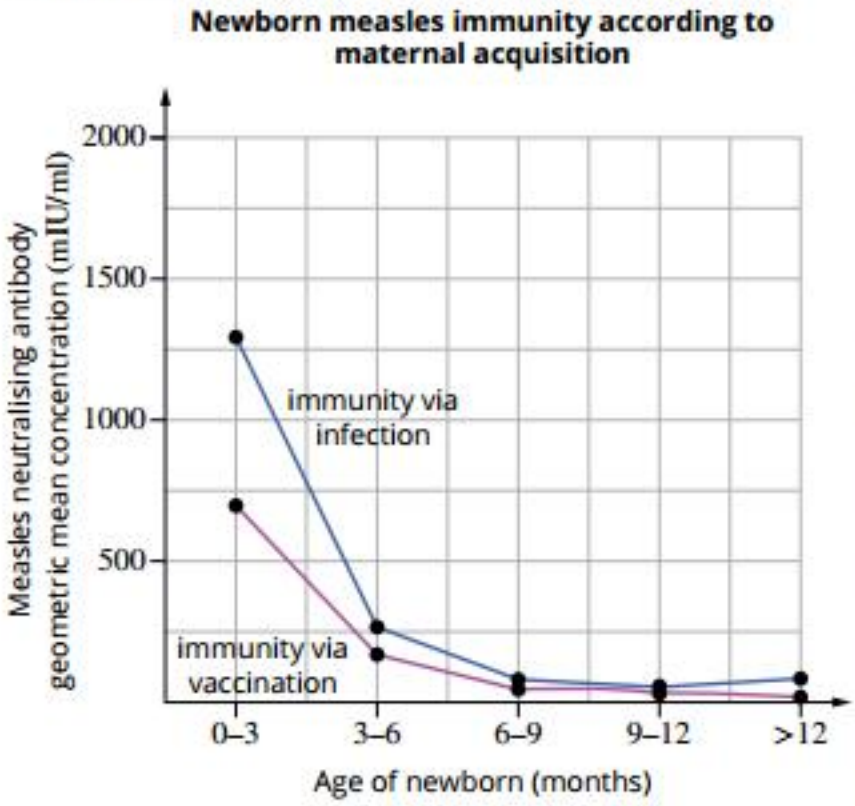
37 Routine measles vaccinations began in France in 1983. In 2005, a study was undertaken to examine the concentration of measles antibodies in French women of childbearing age. The aim was to discover the effect of immunisation on maternal antibody levels. Antibody levels must exceed 200mIU mL^{-1} for protection to be maintained.

The results of this study are shown in the graph.



a Determine the effect of vaccination on long-term antibody levels. Explain your findings.

Following the examination of the adult women, a further study to examine the effects on newborn babies and their antibody levels of the vaccination program was undertaken. The results are shown in the graph.



b Evaluate how the vaccination program affected maternal antibody presence in newborns.

c The first dose of MMR vaccine is routinely given in Australia at 12 months of age. Predict the possible consequences of this in light of the data collected in France and the decline in childhood vaccination rates in Australia.

38 In 2004, a new influenza strain in the H5N1 family of viruses arose in birds in Asia. 'Bird flu' caused worldwide concern, as it became clear that the virus could move from birds to humans. Strict quarantine of anyone showing symptoms contained the disease, but there were still significant numbers of fatalities. Many of the fatalities occurred as a result of acute respiratory distress syndrome (ARDS). ARDS is severe damage to the lungs caused by excessive release of cytokines, which causes many lung cells to undergo apoptosis.

a The stimulation of large numbers of cytokine receptors in lung cells results in the triggering of apoptosis in otherwise healthy cells, by the extrinsic pathway. Explain why lung cells are exposed to many cytokines during an influenza infection.

b The table shows the incidence of bird flu in humans and the number of fatalities up to December 2006.

Country	Time frame	Number of cases	Number of fatalities
Azerbaijan	Feb 06 – Mar 06	8	5
Cambodia	Jan 05 – Mar 06	6	6
China	Oct 05 – Apr 06	18	12
Egypt	Mar 06 – Apr 06	12	4
Indonesia	Jul 05 – Mar 06	32	24
Iraq	Jan 06 – Jan 06	2	2
Thailand	Jan 04 – Nov 05	22	14
Turkey	Dec 05 – Jan 06	12	4
Vietnam	Dec 03 – Nov 05	91	42

i Hypothesise with justification where bird flu might have originated.

ii In all major airports, anyone coming from a country where bird flu had occurred was screened for a raised temperature and, if the result was positive, the person was immediately quarantined. For typical strains of influenza, 9–13% of infected people die. Using the data in the table, explain why such stringent precautions against the spread of bird flu were implemented.

iii Following the outbreak, there was a large research effort to create a successful vaccine, and people in affected areas were encouraged to be vaccinated. Explain how a large-scale vaccination program reduces the risk of a major outbreak of a communicable disease.

c The table shows the distribution of deaths due to bird flu, by age and sex.

Age group (years)	Deaths	
	Male	Female
<5	13	8
5–9	19	13
10–19	18	31
20–29	18	27
30–39	17	16
40–49	5	6
>50	6	5

i Draw a graph using the data in the table.

ii Explain limitations in the data displayed that may result in it being misleading.

iii Explain whether the data suggests that either sex is more susceptible to bird flu.

iv The data given is from a WHO (World Health Organization) report. WHO does not collect its own data. It relies on self-reporting from affected countries. Comment on the reliability of the data presented.

39 Rabies is a serious disease caused by a virus. It infects many animals, including dogs, but does not occur in Australia. After infection, the reproduction of the virus is very slow and the disease is also slow to become symptomatic. Australia's quarantine laws restrict the movement of dogs into the country. These laws were contravened in 2015 when US actor Johnny Depp brought two dogs into Australia.

a Predict the effects of the entry of rabies into the Australian native animal population. Justify your prediction.

b When individuals are bitten by a potentially rabid animal overseas, a treatment regime is initiated. This regime begins with an initial injection of immunoglobulins followed by a vaccination series.

i Explain how the first injection differs from the later series of vaccinations.

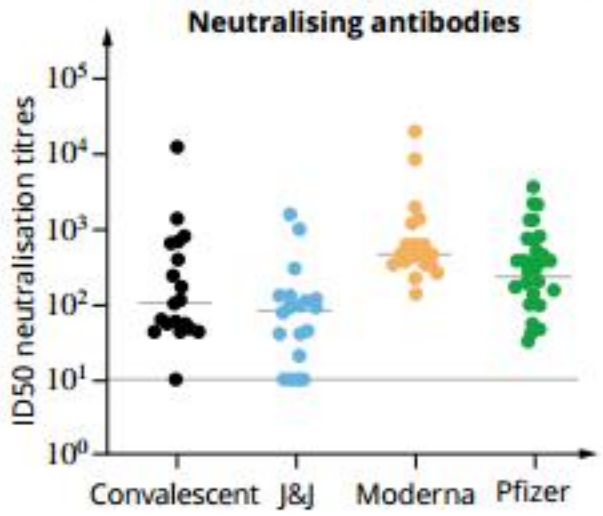
ii State the type of immunity provided by each injection.

iii Suggest why the first injection is given.

iv Propose why the later vaccinations are also required.

40 A study was undertaken to evaluate the effectiveness of three frequently used vaccines for COVID–19. The level of neutralising antibodies present in blood after participants received one of these vaccines was compared with the level in individuals who had been naturally infected with the virus and had recovered (referred to as 'convalescent' in the graph below). The results of the study are shown in the graph below. Black horizontal lines represent the median.

Consider the data and evaluate the three vaccines.



41 Design an experiment that could be performed to test which method of disease control was the most effective against airborne respiratory viruses.

42 According to the WHO (World Health Organization), 'Microbiologically contaminated drinking water can transmit diseases such as diarrhoea, cholera, dysentery, typhoid and polio and is estimated to cause approximately 505 000 deaths a year'.

Investigate methods that can be used to treat water and decide which you would recommend to people living in developing countries. Justify your decision.

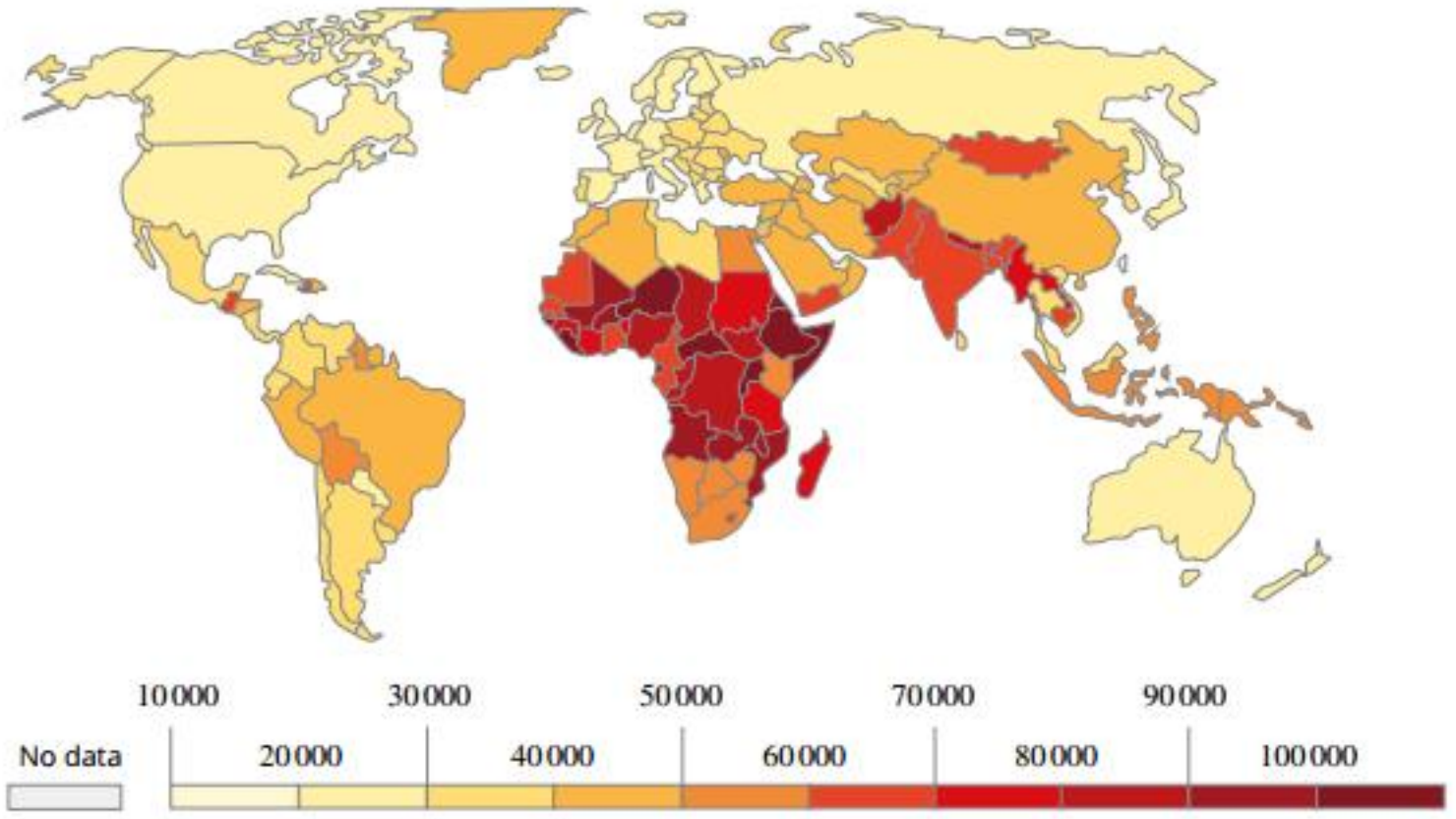
43 During the height of the COVID–19 pandemic, many people were concerned about walking behind joggers. Research the transmission of COVID–19 and justify whether these concerns were logical and valid. Propose some ways that walkers could alleviate their concerns.

44 Avian influenza (or bird flu) is a highly contagious disease that has devastated poultry flocks and wild bird populations. Although the disease mostly affects birds, it is known to infect some mammals. Outbreaks of avian influenza pose a risk to farmers' livelihoods, food security and human health. Research the following:

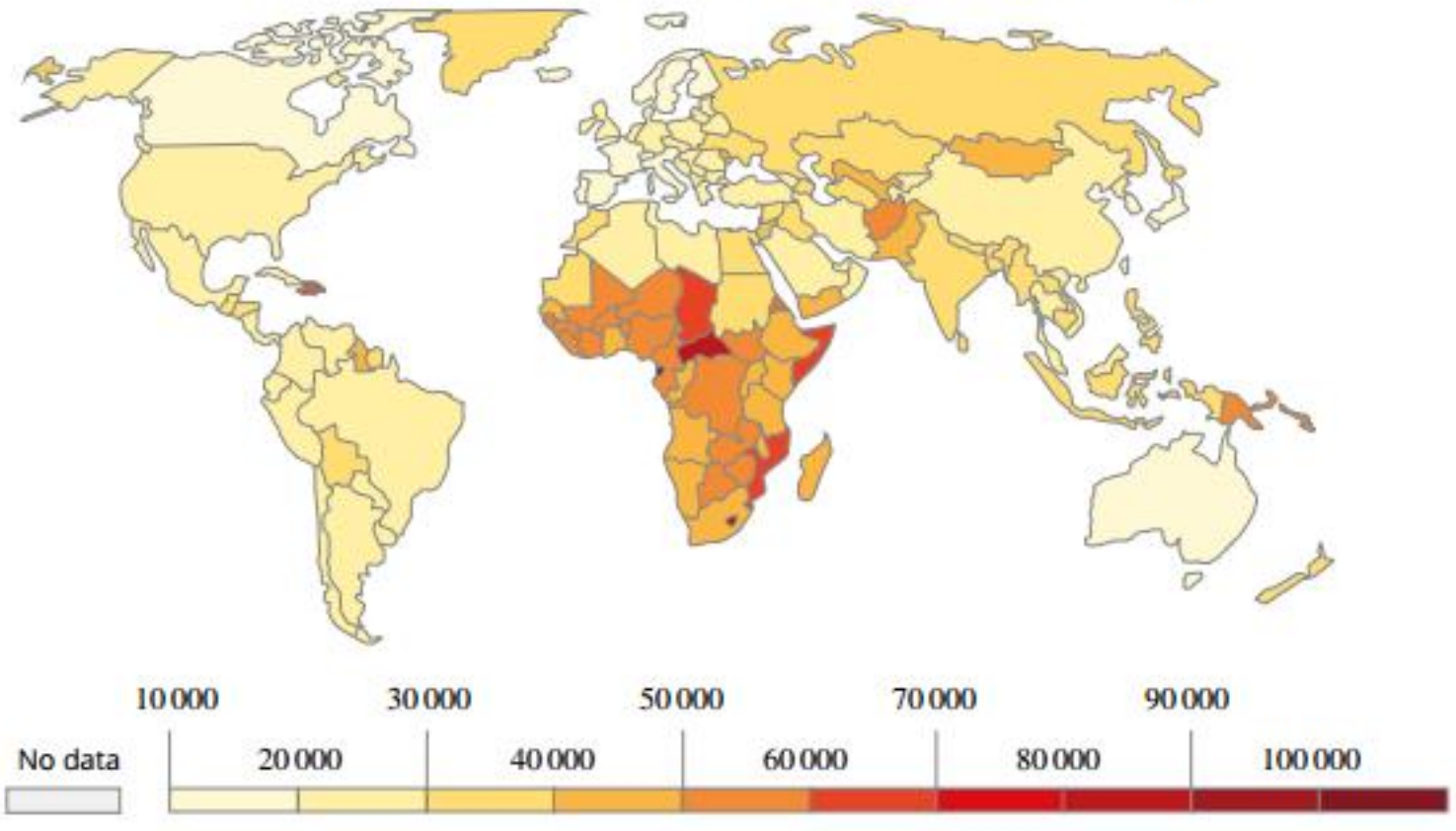
- **a** What does this flu do to birds?
- **b** Why have these outbreaks caused so much harm to other animal populations, farmers' livelihoods, and the food trade?
- **c** Why are scientists concerned about this?
- **d** Have there been any cases within Australia recently?
- **e** Are all strains the same in terms of virulence?
- **f** What can be done to stop the spread of bird flu?

45 The following two charts identify the DALYs (disability-adjusted life years) for every 100 000 people in each country in 1990 and 2019. DALYs measure the total burden of disease. That is, the years of life lost due to premature death and years lived with a disability. One DALY is equal to one lost year of healthy life.

Burden of disease 1990, DALYs per 100 000 people per country



Burden of disease 2019, DALYs per 100 000 people per country



a Compare and contrast the two charts and highlight the differences.

b What are possible reasons for the differences between the two charts?

Practical manual

Practical investigations support the development of science inquiry skills and provide you with hands-on experience to deepen your understanding. The following science inquiry investigations offer opportunities for you to engage with practical applications of scientific concepts. These skills involve identifying and posing questions and working to answer them, evaluating claims, investigating ideas, solving problems, reasoning, drawing valid conclusions and developing evidence-based arguments.

Through these investigations, you will practise writing research questions, planning and conducting experiments, recording information and reflecting on your processes. You will learn to process, analyse and interpret evidence, evaluate conclusions and claims critically and communicate your findings effectively.

Syllabus subject matter

Unit 1: Cells and multicellular organisms

Practical investigation 1: Investigate the effect of surface area-to-volume ratio on rate of diffusion

- Investigate the effect of surface-area-to-volume ratio on rate of diffusion.

Practical investigation 2: Observing cells

- Use a light microscope or photographs to:
  - view tissues from the respiratory, circulatory, excretory, digestive and/or plant systems
  - compare epithelial, connective, muscle and nervous tissues
  - calculate total magnification and field of view.

Practical investigation 3: Investigate the effect of temperature on the reaction rate of enzymes

- Investigate the effect of temperature/pH/substrate concentration on the reaction rate of different enzymes.

Unit 2: Maintaining the internal environment

Practical investigation 4: Investigate adaptations of plants involved in transpiration and gas exchange

- Investigate:
  - factors affecting the rate of transpiration in different plants
  - adaptations that allow for efficient nutrient and/or gas exchange in plants or animals.

Practical investigation 5: Investigate the effect of antimicrobial agents on bacteria

- Investigate the effect of an antimicrobial agent on the growth of a microorganism (via the measurement of zones of inhibition) in either a laboratory or virtual context.

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Investigate the effect of surface area-to-volume ratio on rate of diffusion

Forming and Finding

Aim

To investigate the research question: Does the surface area to volume ratio of cells impact their ability to efficiently transport materials across the cell membrane?

Rationale (scientific background to the experiment)

All cells are surrounded by a cell membrane. The cell membrane is a semipermeable barrier that controls the movement of substances into and out of the cell. Movement across the cell membrane is two-directional, and occurs via diffusion, osmosis or active transport.

Generally, the larger the volume of a cell, the larger the surface area. Surface area-to-volume ratio (SA : V) is a measure of these two factors combined. Smaller cells usually have a larger surface area compared to their volume. This allows cells to move molecules across their cell membranes in an efficient manner. It also explains why single-celled organisms are limited in their size.

The 'pink agar' cubes are models of cell size. They have been prepared using sodium hydroxide and phenolphthalein. Phenolphthalein is an indicator that is pink in alkaline solutions and turns colourless in neutral and acidic solutions. The pink agar turns clear in the presence of sulfuric acid, which is evidence of diffusion.

Timing

60 minutes

Materials

- 3 'pink agar' cubes of the following dimensions: 1 cm^3, 2 cm^3, 3 cm^3
- 100 mL of 0.1 mol L^{-1} sulfuric acid
- cutting board and knife
- ruler with millimetre increments
- plastic spoon
- 250 mL glass beaker
- paper towelling
- timer

Method

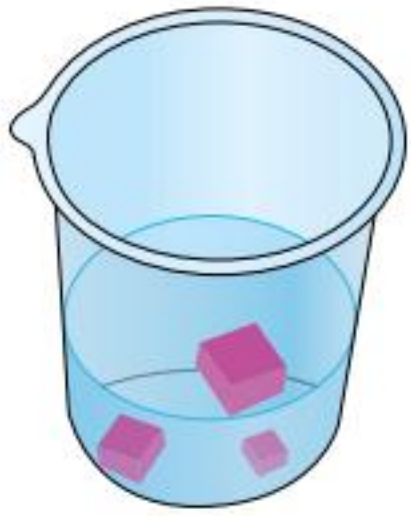
Risk assessment

Assessment of risks include chemical hazards and physical hazards. Before you commence this practical investigation, you must conduct a risk assessment.

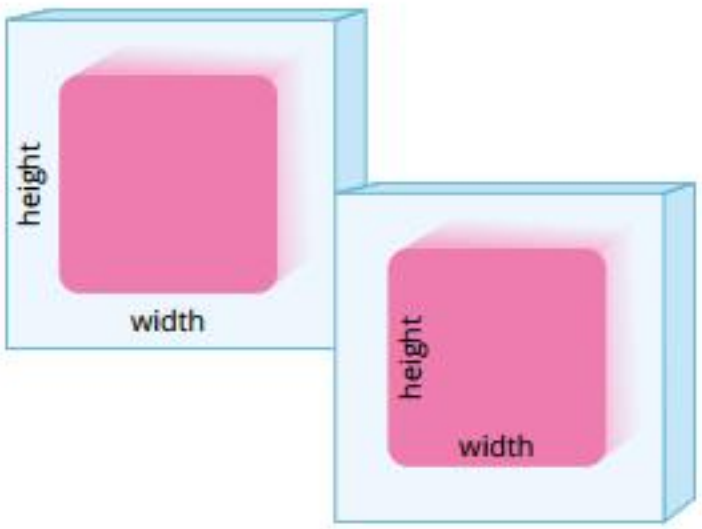
➤ Reflect and check the materials and method for risks

- ☐ Consider and account for risks/ethical issues/ environmental issues in the materials and method.
- ☐ Conduct a risk assessment, completing the template in your Skills and Assessment book or downloading it from your eBook

1. Put on disposable gloves, a lab coat and safety glasses.
2. Gently place each of the three agar cubes in the beaker and cover them with sulfuric acid.



3. Set the timer for 10 minutes.
4. Every 2 minutes, gently turn the agar cubes to ensure even exposure to the acid.
5. At the end of the 10 minutes, gently remove the agar cubes with the spoon and blot onto paper towel to remove excess acid.
6. Cut each cube open and measure the height and width of the remaining pink prism. Assume that the length is the same as the height measurement. It is important to work efficiently at this point as diffusion will continue to occur.



7 Calculate the surface area to volume ratio of each cube.

8 Calculate the rate of diffusion in each cube.

Variables

i Independent: the volume of the cube

ii Dependent: the rate of diffusion

iii Controlled: concentration of sulfuric acid, temperature, time of exposure to acid, amount of stirring

Analysing

Raw data

1 Complete the following table. In the final column, calculate the SA:V so that the volume ratio is 1.

Length of each side of cube (cm)	Surface area (SA) (cm^2)	Volume (V) (cm^3)	SA:V	SA:V, where V is 1
10	$6 \times 10^2 = 600$	$10^3 = 1000$	600:1000	0.6:1
3				
2				
1				
0.1				

Note: 10 cm and 0.1 cm are for comparison only.

Guide to calculations:

- surface area = length × width × number of sides
- volume = length × width × height

Processed data

2 Calculate the percentage of each block that has been reached by the diffusion of acid.

Cube	Dimensions of coloured prism remaining (length, width, height) (cm)	Volume of coloured prism remaining (X) (cm^3)	Volume of whole cube (Y) (cm^3)	Volume of uncoloured portion (Y − X) (cm^3)	% of cube uncoloured $(\frac{Y-X}{Y} \times 100)$
3 cm					
2 cm					
1 cm					

➤ Reflect and check that your data analysis demonstrates these characteristics

- ☐ Effective investigation of phenomena is demonstrated by the collection of sufficient and relevant raw data.
- ☐ Accurate application of algorithms, visual and graphical representations of data is demonstrated by appropriate processing and presentation of data to aid the analysis and interpretation of data.

Analysis

3 Create a new table that shows the relationship between SA:V and percentage of cube uncoloured for each cube.

Compare the percentages of cube uncoloured that you obtained with those of three other groups.

- **a** Explain why the results should be similar for each group.
- **b** If a group did not obtain similar results, suggest a reason for this.

4 Identify two potential errors encountered in the procedure. Suggest how these could be minimised if the procedure were modified.

➤ Reflect and check that your analysis demonstrates these characteristics

- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough and appropriate error analysis.
- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough identification of relevant trends, patterns and relationships.
- ☐ Insightful and valid interpretation of evidence is demonstrated by drawing a valid and defensible conclusion based on the analysis.

Improvements

6 If you were to repeat this experiment, identify the steps that you would do differently. Consider how you could:

- **a** change the methodology
- **b** improve your technique
- **c** reduce error and uncertainty.

Extension

7 If a single-celled organism, such as an amoeba, were to split in half, thus reducing the volume of each compared to the original, what would happen to the SA:V ratio of each new cell compared to the original?

8 In general, motile animal cells are significantly smaller than plant cells. Using the evidence collected in this practical investigation, suggest why.

9 Investigate the cells that line the small intestine. Name the extensions that increase the SA:V ratio. Explain the purpose of these cells in relation to the role of the small intestine.

➤ Reflect and check that your evaluation demonstrates these characteristics

- ☐ Critical evaluation of processes is demonstrated by a discussion of the reliability and validity of the experimental process supported by evidence such as the quality of the data (as quantified in the error analysis).
- ☐ Critical evaluation of the conclusion is demonstrated by a discussion of the veracity of the conclusions with respect to the error analysis and limitations or sufficiency of the data.
- ☐ Insightful evaluation of processes and conclusions is demonstrated by a suggestion of improvements or extensions to the experiment which are logically derived from the analysis of the evidence.

Interpreting and Evaluating

Conclusion

1 State the relationship between surface-area-to-volume ratio and rate of diffusion.

2 Outline the data that you have collected that supports this conclusion.

3 Explain why cells are limited in their ability to grow larger over time.

Evaluation

4 Identify at least one assumption that this practical investigation makes about the shape of cells.

5 Explain whether the potential errors you identified above had a significant effect on your conclusions. In other words, do you consider the level of uncertainty caused by the potential errors reasonable?

Observing cells

Forming and Finding

Aim

To investigate the research question: Can tissues from the respiratory, circulatory, excretory, digestive, and/or plant systems be distinguished using a light microscope, and how do these cells compare?

Apply skills in calculating total magnification and field of view be used to distinguish qualitative differences in cells.

Rationale (scientific background to the experiment)

Cells are the basic building blocks of life. Whether it is a unicellular organism or a complex multicellular organism, all cells have some common features. At the most basic level this includes a cell membrane, cytoplasm, DNA and ribosomes.

Cells are generally classified broadly into prokaryotes and eukaryotes. Eukaryotes are often grouped as either plant or animal cells. In this practical investigation, you will view a range of plant and animal cells. Consider how the cells are both similar and specialised to allow each particular cell type to perform a specific role within the organism. You will also calculate the total magnification and field of view to determine cell size.

Timing

60 minutes

Materials

- light microscope
- microscope lamp
- micrometer grid
- teat pipette
- toothpick
- mounted needle
- small beaker of water
- white tile
- scalpel
- glass microscope slides and coverslips
- paper towel
- onion segment
- small piece of banana
- iodine stain in dropper bottle
- tweezers
- sample *Elodea* plant
- prepared slides of human cheek cells, human blood, mammalian nerve cell/s, and any other cell types available.

Method

Risk assessment

Assessment of risks include chemical hazards and physical hazards. Before you commence this practical investigation, you must conduct a risk assessment.

➤ Reflect and check the materials and method for risks

- ☐ Consider and account for risks/ethical issues/ environmental issues in the materials and method
- ☐ Conduct a risk assessment, completing the template in your Skills and Assessment book or downloading it from your eBook

Part A: Preparing slides

Onion cells

1 Cut off a thin piece of onion.

2 Use the tweezers to peel a very thin layer of epidermis (this looks like tissue paper).

3 Place the sample on a clean glass slide and flatten it as much as possible. Add a single drop of iodine.

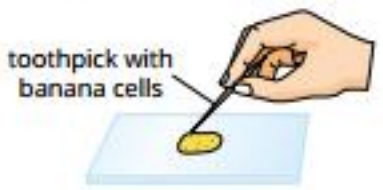
4 Gently lower the coverslip onto the slide, using a mounted needle. Try to minimise any air bubbles. Blot any excess stain at the edge of the coverslip as required.

5 Set the sample aside until you are ready to view it under the microscope.

Banana cells

6 Smear some banana on the toothpick.

7 Put the banana cells on a clean glass slide.



8 Add a single drop of iodine solution and cover with a glass coverslip.

9 Blot any excess stain at the edge of the coverslip as required.

10 Set the sample aside until you are ready to view it under the microscope.

Elodea cells

11 Select a small, thin leaf from the *Elodea* plant.

12 Place the leaf gently onto a glass slide and cover it gently with a coverslip.

13 Set the sample aside until you are ready to view it under the microscope.

Part B: Viewing and drawing slides under the light microscope

14 Set up your microscope on the workbench.

15 Calculate the field of view diameter using the ×4, ×10 and ×40 objective lenses.

16 View each of the slides (those that you prepared and also the pre-prepared ones).

17 Sketch two or three cells accurately, showing the position of the cells in relation to each other. Try to select cells that are not overlapping one another.

18 Include the magnification and a scale of size for each sketch.

19 Note and label the visible organelles within each cell. For the slide of human blood, try to identify the different cell types.

Analysing

Raw data

1 Complete the table.

Microscope magnification	Field of view diameter

Processed data

2 Sketch two or three cells for each slide. For each diagram, include: slide title, magnification, scale, labelled organelles.

➤ Reflect and check that your data analysis demonstrates these characteristics

- ☐ Effective investigation of phenomena is demonstrated by the collection of sufficient and relevant raw data.
- ☐ Accurate application of algorithms, visual and graphical representations of data is demonstrated by appropriate processing and presentation of data to aid the analysis and interpretation of data.

Analysis

3 Explain why it is important that the specimens used to prepare a slide are as thin as possible.

4 List any organelles you did not see. Explain why you did not see these organelles.

5 Plant cells often contain chloroplasts. Suggest why chloroplasts were present in the *Elodea* cells but absent from the banana and onion.

6 Account for why chloroplasts tend to be found around the outer edge of the *Elodea* cells.

7 Suggest why bananas show a large number of leucoplasts in the cells of the fruit.

8 Suggest how the shape of a typical nerve cell (or neuron) enables it to communicate messages within the nervous system.

9 Mature human red blood cells lack a nucleus. Propose why. Summarise the role of red blood cells within the body.

➤ Reflect and check that your analysis demonstrates these characteristics

- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough and appropriate error analysis.
- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough identification of relevant trends, patterns and relationships.
- ☐ Insightful and valid interpretation of evidence is demonstrated by drawing a valid and defensible conclusion based on the analysis.

Interpreting and Evaluating

Conclusion

1 Name the visible features that were common to all cells viewed.

2 Summarise the main ways that plant cells are different from animal cells.

Evaluation

3 Explain why it is necessary to look at multiple cell types before you make generalisations about cells and organelles.

4 Explain whether the potential errors you identified above had a significant effect on your conclusions. In other words, do you consider the level of uncertainty caused by the potential errors reasonable?

Improvements

5 If you were to repeat this experiment, identify the steps that you would do differently. Consider how you:
- **a** might change the methodology
- **b** might improve your technique
- **c** could reduce error and uncertainty.

Extension

6 There are a variety of white blood cells found in human blood. Research the name and role of two specific types of white blood cells.

7 Search the internet for electron micrographs of various cells. Print out the micrographs and label the organelles.

➤ Reflect and check that your evaluation demonstrates these characteristics

- ☐ Critical evaluation of processes is demonstrated by a discussion of the reliability and validity of the experimental process supported by evidence such as the quality of the data (as quantified in the error analysis).
- ☐ Critical evaluation of the conclusion is demonstrated by a discussion of the veracity of the conclusions with respect to the error analysis and limitations or sufficiency of the data.
- ☐ Insightful evaluation of processes and conclusions is demonstrated by a suggestion of improvements or extensions to the experiment that are logically derived from the analysis of the evidence.

PRACTICAL INVESTIGATION 3

Investigate the effect of temperature on the reaction rate of enzymes

Forming and Finding

Aim

To investigate the research question: How does temperature affect the reaction rate of enzymes, specifically the action of the enzyme bromelain on gelatin?

Rationale (scientific background to the experiment)

Pineapple contains a group of proteases called bromelain. Most chefs know not to include fresh pineapple juice in gelatine desserts because the enzyme digests the protein gelatine, and the gelatine will not set (or gel). Bromelain is used in meat tenderisers, in manufacturing precooked cereals, in some cosmetics and in the treatment of inflammation. Bromelain is measured in GDU (gelatine digesting units).

The protein gelatine is obtained by boiling the skin, tendons and ligaments of animals. As a result, it contains various amino acids and collagen. Collagen is a quaternary protein that is involved in maintaining the structure of a cell. It forms molecular cables to strengthen tendons and wide sheets to protect the skin and internal organs of our body. Unlike gelatine, collagen is not soluble in water. When a gelatine solution cools, it forms a matrix that traps water molecules. When the gelatine is digested by the enzyme bromelain, the trapped water molecules are released.

Like all enzymes, bromelain is a protein. The shape of the active site on the enzyme is specific to the substrate it binds. Any variation to this shape affects the efficiency of the enzyme. High temperatures cause enzymes to change their shape (denature), affecting the ability of the substrate to bind to the active site.

The optimum pH range for bromelain is 4.5–7.5 and its optimum temperature is 35–45°C. The maximum operating temperature for industrial applications (reaction time ≤4 h) is 50°C. Bromelain is stable at pH 3–6 and at temperatures up to 60°C.

Timing

45 minutes plus 15 minutes next day for recording data

Material

- 15 mL fresh pineapple juice (blended and strained pineapple)
- 15 mL canned pineapple juice (blended and strained canned pineapple)
- 15 mL distilled water
- gelatine jelly packet (120 mL per group)
- 3 disposable 3 mL pipettes
- 12 test-tubes
- 2 test-tube racks
- test-tube holder
- 3 large beakers (to hold test-tubes)
- ice
- thermometer
- hot plate
- safety mat
- permanent marker

Method

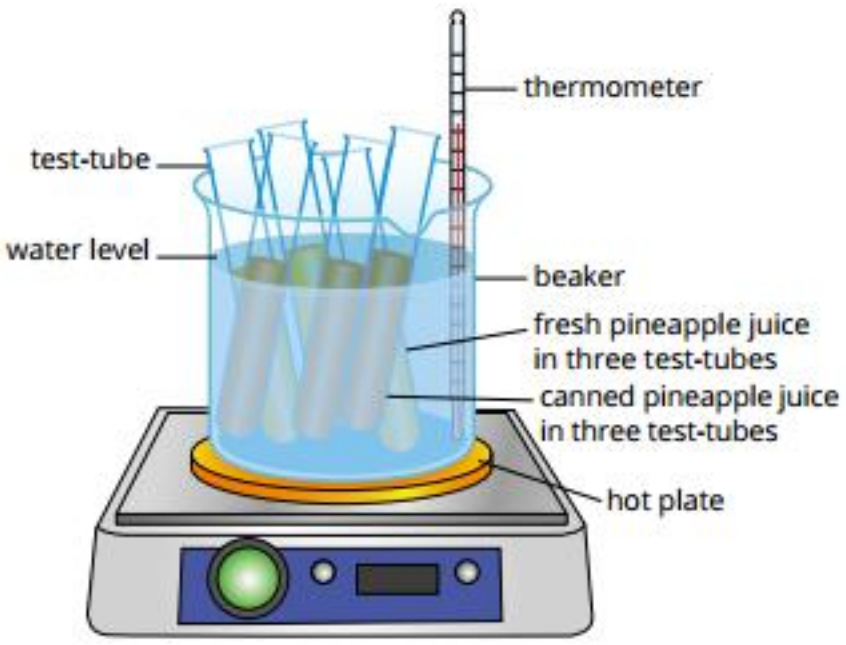
Risk assessment

Assessment of risks include chemical hazards and physical hazards. Before you commence this practical investigation, you must conduct a risk assessment.

➤ Reflect and check the materials and method for risks

- ☐ Consider and account for risks/ethical issues/ environmental issues in the materials and method
- ☐ Conduct a risk assessment, completing the template in your Skills and Assessment book or downloading it from your eBook

1. Make up the gelatine jelly according to the packet instructions. Put the jelly aside until step 11.
2. Number the test-tubes 1–12.
3. Use a disposable pipette to transfer:
   - 3 mL of fresh pineapple juice to test-tubes 1–5
   - 3 mL of canned pineapple juice to test-tubes 6–10
   - 3 mL of distilled water to test-tubes 11 and 12.
4. Place test-tubes 1, 6 and 11 in a beaker of ice and water. Record the temperature.
5. Place test-tubes 2, 7 and 12 in the test-tube rack at room temperature. Record the temperature.



6 Place the remaining test-tubes in the beaker of water with a thermometer.

7 Heat the beaker containing test-tubes slowly on the hot plate.

8 At 40°C, remove test-tubes 3 and 8 from the beaker using the test-tube holder, and allow them to cool in the test-tube rack.

9 At 60°C, remove test-tubes 4 and 9 from the beaker using the test-tube holder, and allow them to cool in the test-tube rack.

10 At 80°C, remove test-tubes 5 and 10 from the beaker using the test-tube holder, and allow them to cool in the test-tube rack.

11 Add 10 mL of gelatine mixture to all 12 test-tubes containing the treated pineapple solutions and the distilled water at room temperature.

12 Cover the test-tubes with plastic wrap and allow them to cool in the fridge overnight.

13 Record the level of solidification or liquidity of the contents. Note whether or not gelatine is present and measure the amount of any liquid present in the test-tubes.

Variables

i Independent: temperature, type of pineapple juice

ii Dependent: solidification of gelatine

iii Controlled: amount of pineapple juice, amount of gelatine, time for gelatine to set

Analysing

Raw data

1 Complete the table to record the effect of temperature on bromelain function.

Temperature (°C)	Gelatine present			Volume of liquid present (mL)		
	Fresh pineapple	Canned pineapple	Distilled water	Fresh pineapple	Canned pineapple	Distilled water
0						
20						
40						
60						
80						

Processed data

2 Create a graph showing the effect of temperature on bromelain function for both fresh and canned pineapple juice.

➤ Reflect and check that your data analysis demonstrates these characteristics

- ☐ Effective investigation of phenomena is demonstrated by the collection of sufficient and relevant raw data.
- ☐ Accurate application of algorithms, visual and graphical representations of data is demonstrated by appropriate processing and presentation of data to aid the analysis and interpretation of data.

Analysis

3 Explain why water was used in test-tubes 11 and 12.

4 Explain why a test-tube of water was not heated to 40°C, 60°C and 80°C.

5 Explain why a Bunsen burner was not used to heat the test-tubes directly.

6 Review the results for the canned pineapple juice. Explain why gelatine did, or did not, form.

7 State at what temperature bromelain denatures. Provide evidence from your results to support your answer.

8 Draw a labelled picture that shows what happens at a molecular level when the enzyme bromelain is heated.

9 Compare your results with those from three other groups.

a Explain why the results should be similar for each group.

b If a group did not obtain similar results, suggest a reason for this.

10 Identify a potential error encountered in the procedure. Suggest how it could be minimised if the procedure were modified.

➤ **Reflect and check that your analysis demonstrates these characteristics**
☐ Systematic and effective analysis of evidence is demonstrated by a thorough and appropriate error analysis.
☐ Systematic and effective analysis of evidence is demonstrated by a thorough identification of relevant trends, patterns and relationships.
☐ Insightful and valid interpretation of evidence is demonstrated by drawing a valid and defensible conclusion based on the analysis.

Interpreting and Evaluating

Conclusion

1 Describe how temperature affected the action of bromelain on gelatine.

Evaluation

2 Explain whether the potential errors you identified above had a significant effect on your conclusions. In other words, do you consider the level of uncertainty caused by the potential errors reasonable?

Improvements

3 If you were to repeat this experiment, identify the steps that you would do differently.
Consider how you could:

a change the methodology
b improve your technique
c reduce error and uncertainty.

Extension

4 Investigate two other enzymes that are used in cooking. Find out the optimal pH and temperature ranges at which they operate and explain how this information is used in cooking.

5 Doctors become very concerned if a patient has a prolonged and very high fever. Use your understanding of proteins and their role in the body to explain why.

➤ **Reflect and check that your analysis demonstrates these characteristics**
☐ Critical evaluation of processes is demonstrated by a discussion of the reliability and validity of the experimental process supported by evidence such as the quality of the data (as quantified in the error analysis).
☐ Critical evaluation of the conclusion is demonstrated by a discussion of the veracity of the conclusions with respect to the error analysis and limitations or sufficiency of the data.
☐ Insightful evaluation of processes and conclusions is demonstrated by a suggestion of improvements or extensions to the experiment which are logically derived from the analysis of the evidence.

Investigate adaptations of plants involved in transpiration and gas exchange

Forming and Finding

Aim

To investigate the research question: What are the adaptations of plants involved in transpiration and gas exchange, and how do these adaptations vary in plants from different environments?

Rationale (scientific background to the experiment)

The leaf is the primary site of photosynthesis in plants. In order to carry out photosynthesis, gases and water vapour must be exchanged with the environment. The stomata and guard cells control the movement of these substances into and out of the plant.

Stomata and guard cells also play an important role in osmoregulation in plants. When stomata are open, water vapour exits the plant. The exiting water pulls water up the xylem vessels from the roots, ensuring continuous one-way flow of water from roots to leaves, known as transpiration.

Up to 90% of water loss in plants occurs through transpiration. This significant amount of water loss is a problem for plants living in environments with limited water. Consequently, plants have adapted several structural features that reduce the rate of transpiration, a number of which relate to stomata and guard cells.

You will examine the stomata and leaves from a range of plants adapted to live in various environments to identify structural adaptations in plants.

Timing

60 minutes

Materials

- light microscope
- microscope lamp
- leaves from various plants (e.g. *Eucalyptus*, *Bergenia*, lily)
- clear fingernail polish
- clear sticky tape
- microscope slides
- forceps
- scalpel
- scissors

Method

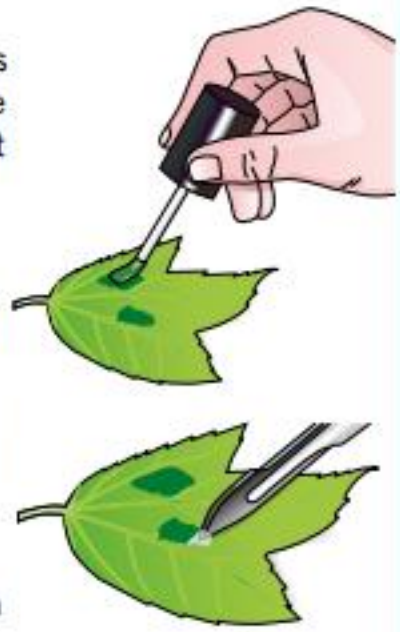
Risk assessment

Assessment of risks include chemical hazards and physical hazards. Before you commence this practical investigation, you must conduct a risk assessment.

➤ Reflect and check the materials and method for risks

- ☐ Consider and account for risks/ethical issues/environmental issues in the materials and method
- ☐ Conduct a risk assessment, completing the template in your Skills and Assessment book or downloading it from your eBook

Part A: Preparing slides



1 Paint three thick 1 cm^2 squares of nail polish on one side of the leaf being studied. Do not paint the polish over the leaf veins.

2 Allow the nail polish to dry.

3 Use the scalpel to carefully lift one corner or side of the dried nail polish. Continue carefully separating the polish from the leaf with the scalpel or forceps.

4 Tape a piece of sticky tape to the patch of nail polish.

5 Tape the peeled nail polish to a microscope slide. Use scissors to cut off any excess tape.

6 Label the slide, indicating the leaf type and location (e.g. *Eucalyptus*, top, or *Bergenia*, bottom).

Eucalyptus, top	

7 Repeat the above steps to make mounts for the remaining nail polish patches, and for the top and bottom sides of each leaf.

Part B: Viewing and drawing slides under the light microscope

1 Set your microscope up on the workbench.
2 Calculate the field of view diameter at ×400 magnification.
3 View each of the slides you have prepared at ×400 magnification. You may need to close the condenser aperture to better observe the stomata and guard cells.
4 Sketch what you observe under the microscope, labelling any stomata, guard cells and epidermal cells.
5 Count the number of stomata in your field of view, and record this in a table. Include stomata in your count only if you can see the entire opening.
6 Repeat for the other two patches you have prepared.

Analysing

Raw data

1 Complete the table.

Leaf species	Epidermal layer (top or bottom)	Number of stomata viewed			Average number of stomata
		Sample 1	Sample 2	Sample 3	

Processed data

2 Sketch the epidermal layer using the ×10 objective lens.
For each diagram, include: slide title, magnification, scale, labelled organelles.
3 Use the two formulae below to calculate the area of the field of view (FOV) and the density of stomata for each leaf. Record these values in the table:

$$\text{FOV area} = \pi \times (\tfrac{1}{2}\,\text{FOV diameter})^2$$

$$\text{stomata density} = \frac{\text{number of stomata}}{\text{FOV area}}$$

Leaf species	Epidermal layer (top or bottom)	FOV diameter (mm)	FOV area (mm^2)	Number of stomata	Stomata density (stomata/mm^2)

➤ Reflect and check that your data analysis demonstrates these characteristics

- ☐ Effective investigation of phenomena is demonstrated by the collection of sufficient and relevant raw data.
- ☐ Accurate application of algorithms, visual and graphical representations of data is demonstrated by appropriate processing and presentation of data to aid the analysis and interpretation of data.

Analysis

4 Explain why it is important that you did not touch the nail polish with your fingers.
5 Explain why you needed to use three samples for each side of each leaf.
6 List the species and leaf side for which you saw stomata and guard cells.
7 The distribution of stomata and guard cells in leaves is a structural adaptation to reduce excessive water loss while still allowing gas exchange to occur. Review the density of stomata and leaves, and account for the distribution patterns you observed based on the plant's known habitat.
8 Did any of the leaves you observed lack stomata entirely? Explain why.

➤ Reflect and check that your analysis demonstrates these characteristics

- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough and appropriate error analysis.
- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough identification of relevant trends, patterns and relationships.
- ☐ Insightful and valid interpretation of evidence is demonstrated by drawing a valid and defensible conclusion based on the analysis.

Interpreting and Evaluating

Conclusion

1 Summarise the relationship between stomata density, stomata location, and plant habitat.

Evaluation

2 Suggest why a higher magnification may not be beneficial for this experiment.
3 Explain whether the potential errors you identified above had a significant effect on your conclusions. In other words, do you consider the level of uncertainty caused by the potential errors reasonable?

Improvements

4 If you were to repeat this experiment, identify the steps that you would do differently. Consider how you:
a might change the methodology
b might improve your technique
c could reduce error and uncertainty.

Extension

5 Search the internet for images of stomata for two of the plant species you observed. Ensure the images have a scale. Print out the micrographs and calculate or measure the size of the stomata.

6 Determine the average rainfall for the area where you live. Suggest which plant species you observed would be best suited to grow in your area.

7 Search the internet for images of stomata on plants that live in deserts. Print out the micrographs, calculate stomata density and explain stomata distribution.

➤ **Reflect and check that your evaluation demonstrates these characteristics**
☐ Critical evaluation of processes is demonstrated by a discussion of the reliability and validity of the experimental process supported by evidence such as the quality of the data (as quantified in the error analysis).
☐ Critical evaluation of the conclusion is demonstrated by a discussion of the veracity of the conclusions with respects to the error analysis and limitations or sufficiency of the data.
☐ Insightful evaluation of processes and conclusions is demonstrated by a suggestion of improvements or extensions to the experiment which are logically derived from the analysis of the evidence.

Investigate the effect of antimicrobial agents on bacteria

Forming and Finding

Aim

To investigate the research question: What are the effects of antimicrobial agents, including antibiotics, on bacterial growth? Specifically, how do antimicrobial agents influence the growth of bacteria as measured by zones of inhibition?

Rationale (scientific background to the experiment)

Like all living organisms, bacteria require nutrients in order to grow and reproduce. Within laboratories, colonies of bacteria can be grown on agar plates–Petri dishes that contain a gelatinous substance. Agar plates can also contain bacterial growth inhibitors, such as antibiotics. Different kinds of bacteria are affected by different antibiotics and growth inhibitors. Scientists can identify bacteria based on the appearance of bacterial colonies, and the type of agar on which they grow.

In addition to agar plates, scientists can use Mastrings or Multodiscs to identify bacteria. Mastrings or Multodiscs are commercially produced discs impregnated with a variety of different antibiotic substances. A Mastring comprises a central ring that has a series of satellite rings coming off it. Each satellite on the Mastring contains a different antibiotic that is identified by either a colour or a letter code or both. When bacterial growth is inhibited, clear zones are visible around the satellite. These clear zones are called inhibition zones. Bacterial growth (or inhibition zones) at each satellite enables scientists to identify antibiotics that can be used to kill bacteria.

Timing

2 × 60 minutes, 48 hours apart

Materials

- 3 sterile, prepared nutrient agar Petri dishes
- broth cultures of *Escherichia coli* (*E. coli*) and *Staphylococcus albus* (*S. albus*)
- 2 sterile swabs in container
- marking pen
- fine forceps
- parafilm strips
- access to incubator set at 30°C
- 2 sterile Mastrings
- disposal bags
- paper towel
- disinfectant
- access to material for washing hands
- Bunsen burner

Method

Risk assessment

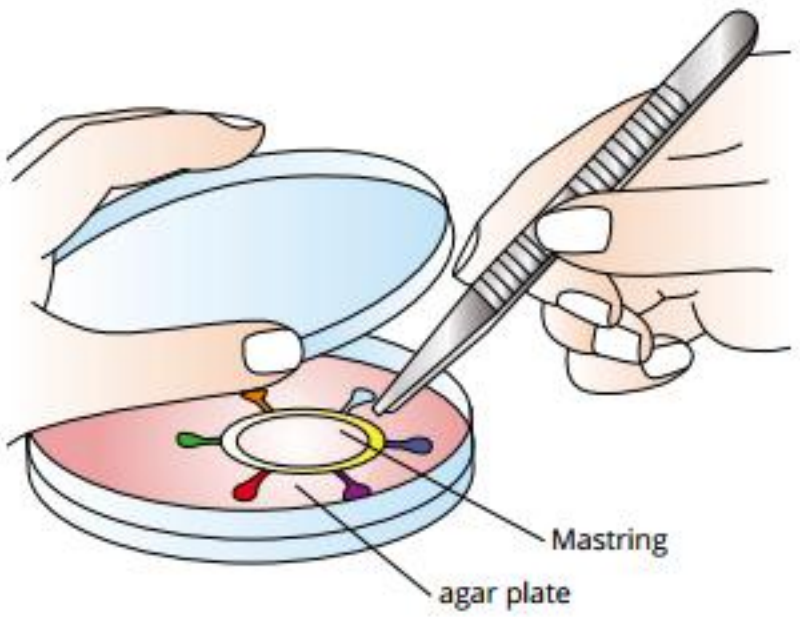
Assessment of risks include chemical hazards and physical hazards. Before you commence this practical investigation, you must conduct a risk assessment.

➤ Reflect and check the materials and method for risks

- ☐ Consider and account for risks, ethical issues and environmental issues in the materials and method
- ☐ Conduct a risk assessment, completing the template in your Skills and Assessment book or downloading it from your eBook

Part A: Day 1

1. Collect three sterile nutrient agar Petri dishes. Keep the lids in place and use a marker pen to label the three dishes 'A', 'B—*E. coli*,' and 'C—*S. albus*'. Use initials or another code to identify your Petri dishes from other students' Petri dishes. Label the underside of the dish only, and write at the perimeter of the dish rather than the middle.
2. Collect a container of each of the two bacteria and two sterile swabs. Your teacher will demonstrate how to prepare a 'lawn culture' of the bacterium. Use the swabs to prepare bacterial cultures of *E. coli* and *S. albus* in Petri dishes B and C, respectively. Immediately replace the lids. Place the used swabs in a disposal bag.
3. Collect two Mastrings. Use fine forceps to place one Mastring in the centre of Petri dish B. Replace the lid. Repeat this procedure for Petri dish C.
   Note: Sterilise the tips of a pair of forceps by flaming in a Bunsen flame. Allow to cool.
   Transfer one Mastring to each of the coated nutrient agar plates, gently pressing the lobes of the disc onto the surface of the plates. Re-sterilise the forceps in between testing different bacterial cultures.



4 Seal each of the Petri dishes with a single layer of parafilm strips.

5 Set the Petri dishes in the incubator. The Petri dishes should be placed upside down, with the agar on the top. This prevents the growth of bacteria being disrupted by drops of condensation.

6 Incubate the Petri dishes at 30°C for 48 hours or at room temperature for 3 days. This will depend on your class timetable and teacher discretion.

7 When all equipment is put away, wipe down your bench with paper towel and disinfectant. Then wash your hands thoroughly.

Part B: Day 2

8 Collect your three Petri dishes for observation. **Do not open the dishes.**

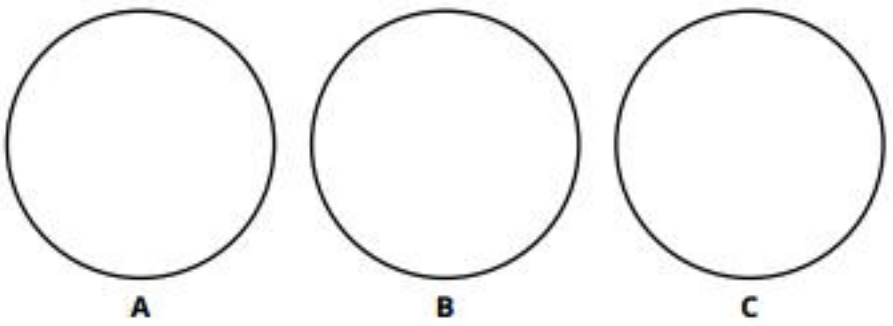
9 Prepare detailed drawings of what you see in each Petri dish. Include appropriate labelling as well as a description of your observations. Measure the inhibition zones around each satellite.

10 Place all Petri dishes in the disposal bag provided. Wash your hands thoroughly.

Analysing

Raw data

1 Copy the circles and space below to record your observations of the Petri dishes. Label each drawing and include a title.



Processed data

2 Look carefully at Petri dishes B and C that include the Mastring. Use your observations to complete the results table.

Record of results

Disc code	Antibiotic on disc	Observations of bacterial growth around disc (diameter of inhibition zone, mm)	
		E. coli	*S. albus*

➤ **Reflect and check that your data analysis demonstrates these characteristics**

- ☐ Effective investigation of phenomena is demonstrated by the collection of sufficient and relevant raw data.
- ☐ Accurate application of algorithms, visual and graphical representations of data is demonstrated by appropriate processing and presentation of data to aid the analysis and interpretation of data.

Analysis

3 Explain the purpose of Petri dish A.

4 Suggest a research question for this investigation.

5 Explain why it is important to ensure the Petri dishes remain sealed.

6 Are all of the inhibition zones the same size? Account for any differences in the size of the inhibition zones.

7 Compare the sensitivity of the bacterial species to the range of antibiotics used.

8 Imagine you are working in a hospital and a patient was infected with *E. coli*. Considering the bacterial sensitivity of both *E. coli* and *S. albus*, suggest which antibiotic you would prescribe and explain why.

9 Identify two potential errors encountered in the procedure. Suggest how these could be minimised if the procedure were modified.

➤ **Reflect and check that your analysis demonstrates these characteristics**

- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough and appropriate error analysis.
- ☐ Systematic and effective analysis of evidence is demonstrated by a thorough identification of relevant trends, patterns and relationships.
- ☐ Insightful and valid interpretation of evidence is demonstrated by drawing a valid and defensible conclusion based on the analysis.

Interpreting and Evaluating

Conclusion

1 Summarise the relationship between bacteria and antibiotics.

Evaluation

2 Explain whether the potential errors you identified above had a significant effect on your conclusions. In other words, do you consider the level of uncertainty caused by the potential errors reasonable? You may like to consider what impact an error would have if this experiment was being used in a hospital to administer antibiotics.

Improvements

3 If you were to repeat this experiment, identify the steps that you would do differently. Consider how you:

- **a** might change the methodology
- **b** might improve your technique
- **c** could reduce error and uncertainty.

4 Describe the shape of the inhibition zones. Suggest a method to measure the area of the inhibition zones.

Extension

5 Search the internet to identify a range of antibiotics that are used to treat common infections.

6 Regular use of antibiotics has resulted in some bacteria developing resistance to antibiotics. Bacteria that are resistant to multiple antibiotics have been termed 'super bugs' in the media. Search the internet to discover how antibiotic-resistant bacteria, or super bugs, are controlled to prevent major outbreaks of infection.

➤ Reflect and check that your evaluation demonstrates these characteristics

- ☐ Critical evaluation of processes is demonstrated by a discussion of the reliability and validity of the experimental process supported by evidence such as the quality of the data (as quantified in the error analysis).
- ☐ Critical evaluation of the conclusion is demonstrated by a discussion of the veracity of the conclusions with respect to the error analysis and limitations or sufficiency of the data.
- ☐ Insightful evaluation of processes and conclusions is demonstrated by a suggestion of improvements or extensions to the experiment which are logically derived from the analysis of the evidence.

Science as a human endeavour

Science as a human endeavour (SHE) features provide essential stimulus for the Research Investigation. These features are structured to develop foundational skills that support relevant Science Inquiry skills through the lens of Science as a human endeavour. This provides a suitable foundation from which you can launch into Research Investigation internal assessment 3 (IA 3) and build proficiency in Assessment objectives 1, 2 and 3. Engaging with the SHE features will provide practise in writing research questions, planning and conducting experiments, recording information and reflecting on your processes. You will learn to process, analyse and interpret evidence, evaluate conclusions and claims critically and communicate your findings effectively.

Syllabus subject matter

Unit 1: Cells and multicellular organisms

Science as a human endeavour 1: Organising complex biological systems

- Appreciate that to make sense of the complexity of biological systems, scientists often divide them into simpler components that are easier to study, but at each level of the biological hierarchy, new properties emerge.

Science as a human endeavour 2: Pluripotent stem cells

- Appreciate that pluripotent stem cells have the potential to be grown into specialised cells that can be used to repair or replace ailing organs and tissues. Advances in technology have allowed scientists to reprogram cells to become pluripotent.

Science as a human endeavour 3: Ethical treatment of animals in research

- Appreciate that the use of animals in research has played an important role in furthering scientific understanding of the structure and function of multicellular organisms. Ethical treatment of animals as sentient beings has been accepted as a global principle in research and the three strategies of replacement, reduction and refinement form the basis of many international guidelines.

Science as a human endeavour 4: Kidneys and healthy bodies

- Appreciate how understanding the anatomy and physiology of different body systems allows medical professionals to predict, diagnose, monitor and treat disease.

Unit 2: Maintaining the internal environment

Science as a human endeavour 5: Cool clothing

- Appreciate how scientists use their understanding of natural systems to develop new technologies. (from Unit 1)
- Appreciate that understanding natural systems can lead to advances in technology and engineering. For example, computer models of human thermoregulation responses, including heat transfer, perspiration, respiration and blood flows, have been developed for use in the design of clothing and environments that aim to protect humans from hyper- and hypothermia.

Science as a human endeavour 6: Enzymes and homeostasis

- Appreciate that living things need to regulate their internal environment so that factors such as pH and temperature are within the tolerance ranges of enzymes that regulate metabolism.

Science as a human endeavour 7: Traditional medicines of First Nations Australians

- Appreciate that for thousands of years, First Nations peoples' knowledges of natural antiseptics and bush medicines have been used to prevent and treat infections.

Science as a human endeavour 8: Scientific advancement through collaboration

- Appreciate that scientific advancement and the development of complex models often requires contribution from multiple individuals across a range of disciplines.

Science as a human endeavour 9: Protecting our borders—Australian biosecurity

- Appreciate that Australia has an advantage over many other countries because its borders are easier to protect against the influx of disease-carrying materials and organisms. However, as global trade and air travel become more prevalent, it is increasingly important for Australia to protect its agriculture industry and environment through quarantine measures. These include surveillance, monitoring, examination and clearance activities and conform to policies and protocols that are based on scientific data and risk analysis.

Science as a human endeavour 10: Vaccination programs

- Appreciate that mass vaccination programs are more successful when informed by disease outbreak models.

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Organising complex biological systems

Scientific approaches to the 'meaning of life' have been to investigate the origin of life (known as 'abiogenesis'), to understand how so many different lifeforms have developed, and to study how living things maintain and reproduce themselves. Some who have grappled with a definition for 'life' have reduced it to replication of DNA for genes to be passed on. This is referred to as a gene-centred view of evolution and is a modern development from Darwin's theory of natural selection. James Watson, who helped discover the structure and function of DNA, is famously quoted as saying 'I don't think we are here for anything. We're just products of evolution'.

Advances in technology

Scientists continually revise, refine and update their thinking as new evidence becomes available. The complexity of living things usually requires simplified investigative approaches. Often, developments in technology go hand-in-hand with the acquisition of further knowledge. Advances in microscopy have been fundamental to our understanding of biological cells, from the earliest observation of dead cork cells by Robert Hooke (1665), with his simple hand-made light microscope, to the sophisticated electron microscopes of today.

The fundamental question of what is alive and what is not was addressed by the cell theory proposed in the 1830s—that living things are composed of cells and cell products. As knowledge progressed, it was understood that cells contain specialised subunits called organelles, each with specific properties to enable their function in the organisation of a cell. Eventually, the interface of research in chemistry, cell biology and technology made it apparent that living things are a multitude of atoms and molecules interacting in highly complex ways. PCR (polymerase chain reaction) technology is just one tool that has accelerated studies in molecular biology. It has been dubbed 'molecular photocopying' because it is used to make many copies of DNA.

Biological hierarchy

Biologists refer to a hierarchy of organisation in multicellular organisms from cells to whole organisms. Each level can be studied independently but with the awareness that there are interactions between these levels as the organism regulates its internal environment. In Chapter 4 you learnt that multicellular organisms can be organised into cells, tissues, organs and systems (Figure 10.1.1).

There are also levels of organisation below cells and above organisms—cells are made from atoms that form molecules and proteins, and organisms are part of higher levels of biological organisation, such as populations, ecosystems and the biosphere (Figure 10.1.2).

At the molecular level, properties of chemicals are predictable. When many molecules associate to form cell organelles, things become more complex. Scientists start with the smallest unit, usually biomolecules, then describe an organelle as a collection of molecules, then group the organelles of a cell together, then similar cells form a tissue, then tissues that coordinate together form an organ, then a system of organs with a common function. The systems work together to form a functioning multicellular organism. The hierarchy is based on size, levels of complexity and function. The complexity of a whole organism is easier to study and discuss when sub-divided into similar groupings, such as a whole cell, or a single organelle inside the cell. At the same time, having a hierarchy is a reminder of the interaction that exists between all the levels. Each step, from molecule to cell to organism, achieves more functionality than the previous level and requires a high degree of coordination.

KEY QUESTIONS

Forming and finding

1 Identify and explain how two technologies have extended biological knowledge.

2 Draw your own flow diagram of the biological levels of organisation, adding an example at each level.

3 Consult a credible source to find out more about abiogenesis.
   - **a** Explain the term 'abiogenesis'.
   - **b** Is your source a primary or secondary source?
   - **c** Record a citation for your source.

4 How do scientists use a hierarchy to help manage the complexity of knowledge about biological systems?

Analysing

5 A biologist extracted and studied chloroplast organelles in isolation in a laboratory.
   - **a** Explain the advantages of doing this.
   - **b** Assess whether this study will provide an accurate picture of a functioning photosynthetic plant cell.

6 Why is it necessary for a complex multicellular organism to have multiple layers of organisation? In your answer refer to a unicellular organism to identify the advantages and limitations for comparison.

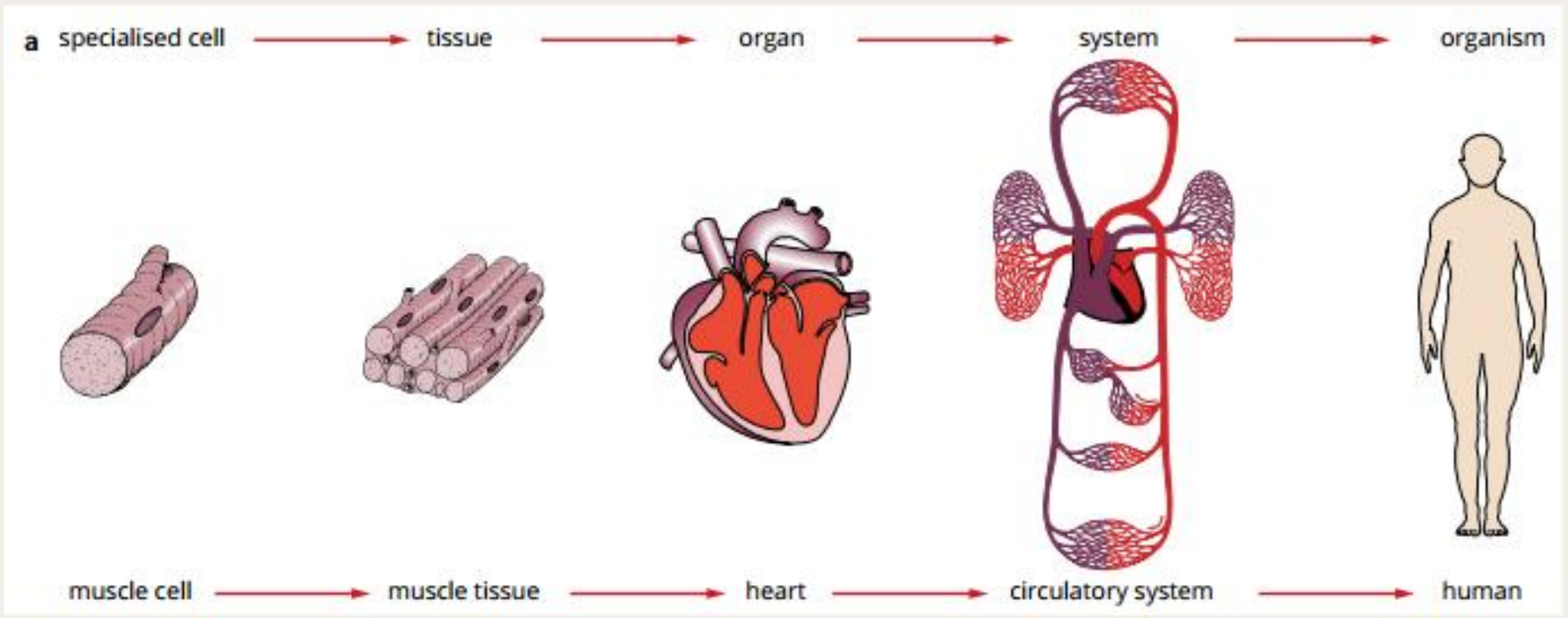


FIGURE 10.1.1 (a) The levels of organisation in a complex multicellular animal: cell, tissue, organ, system and organism. (b) The levels of organisation in a complex multicellular plant: cell, tissue, organ, organ system and organism

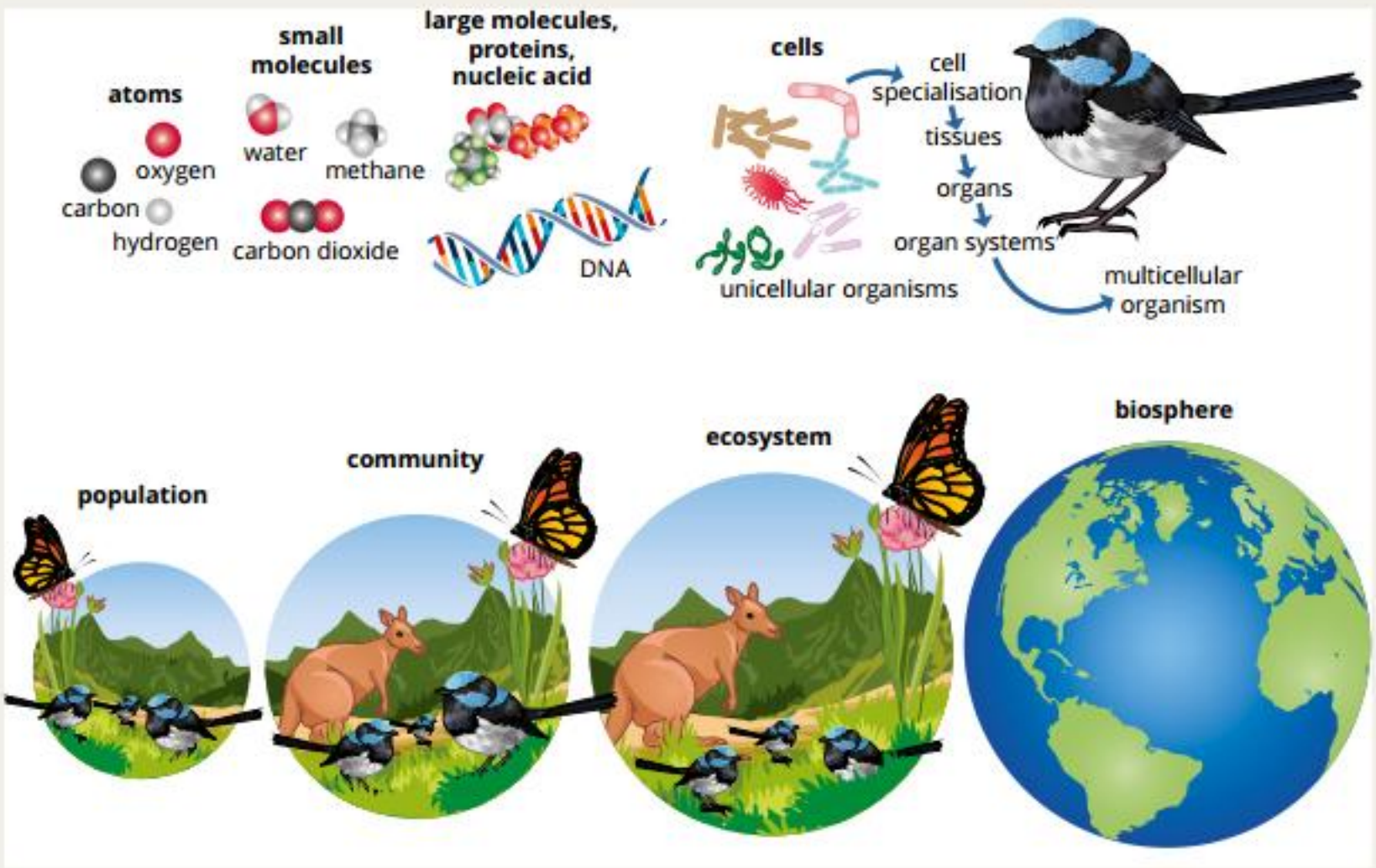


FIGURE 10.1.2 Levels of biological organisation, from (a) atoms to organisms and (b) organisms to ecosystems

Pluripotent stem cells

You will recall from Chapter 4 the types of stem cells and their different levels of potency. Biomedical research has targeted pluripotent stem cells as being the most promising to generate healthy laboratory-cultured tissues and organs to replace damaged parts of a human body. Transplant organs, such as heart and kidney, are in very short supply and the incidence of kidney disease continues to rise, likely linked to the increase in diabetes. Regeneration of damaged heart, liver, lung, nerve and other tissues is theoretically possible with cultured pluripotent cells.

However, ethical considerations had restricted stem cell research because harvesting the pluripotent cells destroyed embryos at the blastocyst stage. Improved harvesting techniques that do not damage the embryo are being developed. Even more promising is the development of induced pluripotent stem cells (iPSCs). iPSCs are adult cells that have been genetically reprogrammed to an embryonic stem cell-like state. This forces the cell to express genes and factors that are characteristic of embryonic stem cells. Although iPSCs by definition are pluripotent stem cells, it is not yet determined if there are clinical differences in the two stem cell types.

The 2012 Nobel Prize in Physiology or Medicine was awarded to two scientists who worked 40 years apart in different countries (Figure 10.2.1). The work of John B. Gurdon first showed that an isolated mature nucleus could be returned to an immature pluripotent state. Later, Shinya Yamanaka discovered, by introducing four particular genes, how to reprogram intact mature cells back into immature stem cells. The resulting iPSCs could develop into mature cell types such as fibroblasts, nerve cells and gut cells. This means pluripotent cells are available without harvesting from embryos.

In 2015, researchers from the University of California, USA, published the results of a study in which they used human iPSCs to grow miniature 'beating hearts'. These were nothing like the four-chambered heart inside your chest, but the cells did form tiny hollow, pulsating chambers rather than simple layers of cells. Previously stem cells were grown in larger wells like that shown in Figure 10.2.2 and only formed flat sheets. Once they had produced the beating microchambers, the researchers successfully tested them for reaction to a drug that changed the contraction rate.

Stem cells are unlikely to be used to produce replacement hearts for people in the near future, but thanks to this new research, the heart microchambers can be used for drug testing and other biomedical research.

KEY QUESTIONS

Forming and finding

1 Distinguish between the following terms for early development stages of a human embryo: zygote, morula and blastocyst.

2 Describe what is meant by an induced pluripotent stem cell (iPSC).

3 Outline a benefit of using iPSCs rather than embryonic stem cells.

4 Explain the benefit of using beating heart microchambers to test drugs for side effects on the heart.

Analysing

5 Use secondary sources to investigate progress since 2015 in the field of iPSC research.
   - **a** Outline one promising development.
   - **b** For your chosen area, predict the impact of iPSC research and development on human health outcomes.
   - **c** Cite your source using the APA 7 referencing method.

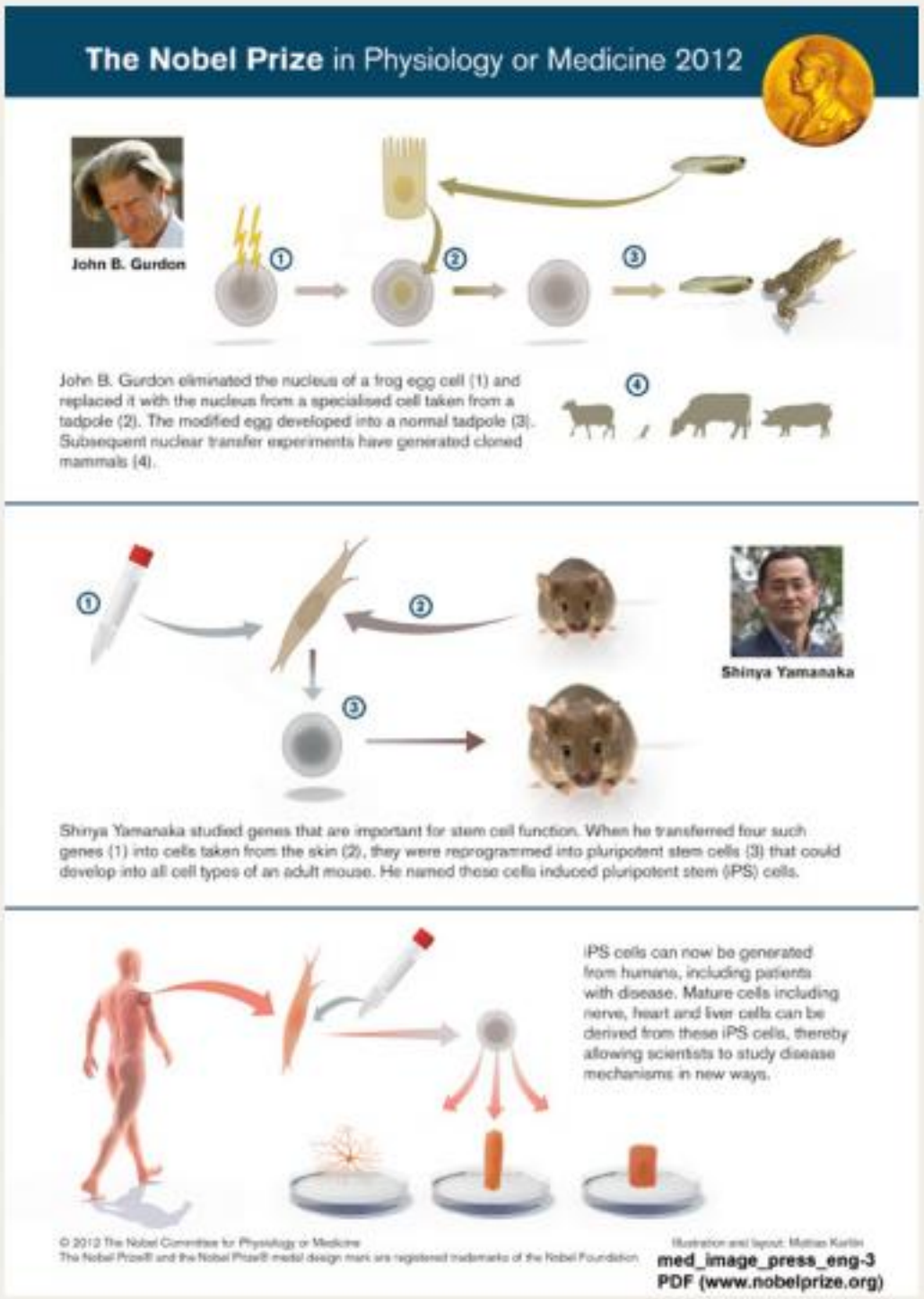

The Nobel Prize in Physiology or Medicine 2012
John B. Gurdon
John B. Gurdon eliminated the nucleus of a frog egg cell (1) and replaced it with the nucleus from a specialised cell taken from a tadpole (2). The modified egg developed into a normal tadpole (3). Subsequent nuclear transfer experiments have generated cloned mammals (4).
Shinya Yamanaka
Shinya Yamanaka studied genes that are important for stem cell function. When he transferred four such genes (1) into cells taken from the skin (2), they were reprogrammed into pluripotent stem cells (3) that could develop into all cell types of an adult mouse. He named these cells induced pluripotent stem (iPS) cells.
iPS cells can now be generated from humans, including patients with disease. Mature cells including nerve, heart and liver cells can be derived from these iPS cells, thereby allowing scientists to study disease mechanisms in new ways.
© 2012 The Nobel Committee for Physiology or Medicine
The Nobel Prize® and the Nobel Prize® medal design mark are registered trademarks of the Nobel Foundation
Illustration and layout: Mattias Karlén
med_image_press_eng-3
PDF (www.nobelprize.org)

FIGURE 10.2.1 The Nobel Prize in Physiology or Medicine 2012 was awarded jointly to John B. Gurdon and Shinya Yamanaka for the discovery that mature cells can be reprogrammed to become pluripotent.

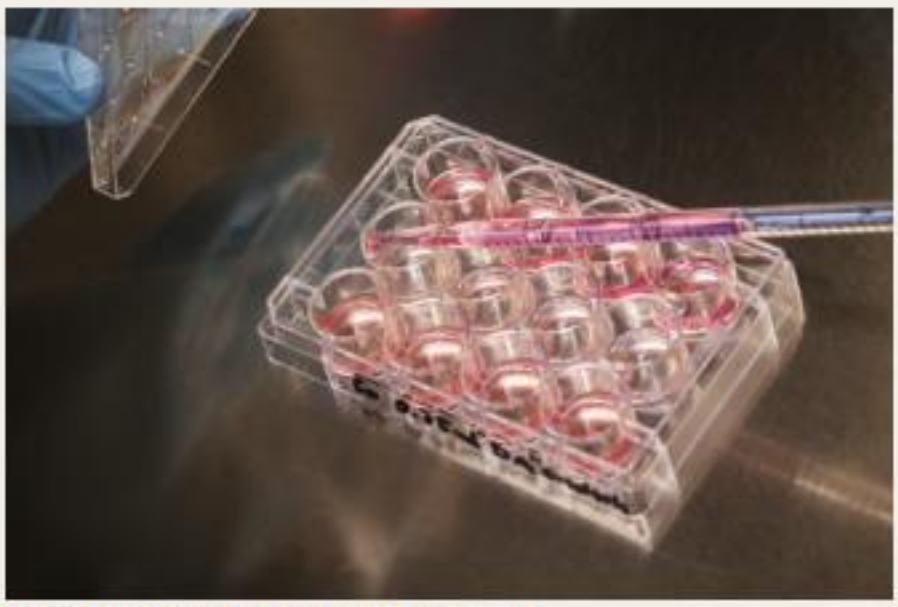


FIGURE 10.2.2 A standard 12-well culture dish containing human stem cells

Ethical treatment of animals in research

From the 4th century BCE, there are records of biomedical research using vertebrate animals as models for understanding human anatomy, physiology, psychology, pharmacology and medical issues. Today, animal-based research usually involves vertebrates, such as cats, rats, mice, frogs, pigs, and sometimes primates, in laboratory settings under carefully controlled conditions. Most animals used in research are specifically bred for use in medical research. The research must be relevant to human or animal health and the studies need to protect the animals' welfare.

Historical perspective

Galen of Pergamon (2nd–3rd century) was a Greek physician who practised in Rome and became a dominant influence on medical theory and practice. Much of his knowledge was gained from animal experiments and dissections, often shared in public. For example, he distinguished seven pairs of cranial nerves, described the valves of the heart, and observed structural differences between arteries and veins. One of his most convincing demonstrations was that arteries carry blood, not air, as had been taught for the previous 400 years. Although this fact seems obvious now, it should be recognised that there was an absence of medical technology and lack of knowledge of human anatomy at the time.

In 12th century Moorish Spain, Ibn Zuhr was a celebrated Arab physician who introduced animal testing to trial surgical techniques before applying them to human patients. Among other achievements, he developed surgical procedures for tracheotomy, excision of cataracts and removal of kidney stones. At the time they were groundbreaking new medical developments.

Some prominent behavioural psychologists developed theories about learning processes by manipulating conditions for live animals in laboratories. Such research has advanced our knowledge of human behaviour and is still widely referred to in educational psychology. Two of the most famous are Ivan Pavlov (1849–1936) and B.F. Skinner (1904–1990):

- Pavlov focused on 'classical conditioning' using dogs to show that repeated experience of a stimulus can lead to anticipating an associated event (Figure 10.3.1a).
- Skinner worked on 'operant conditioning' using reinforcement with rats and pigeons in the Skinner box he developed (Figure 10.3.1b).

Animal protection

In recent times, a different ethical perspective has emerged on animal-based research. Certain animals are now widely regarded as sentient beings—that is they have self-awareness and can experience pain and fear. Some countries have legislation to recognise sentience in animals. In Australia, only the ACT has enacted this type of legislation, with Victoria considering it for the future. The other states, without recognising sentience, have strict animal welfare laws in place for research with live animals.

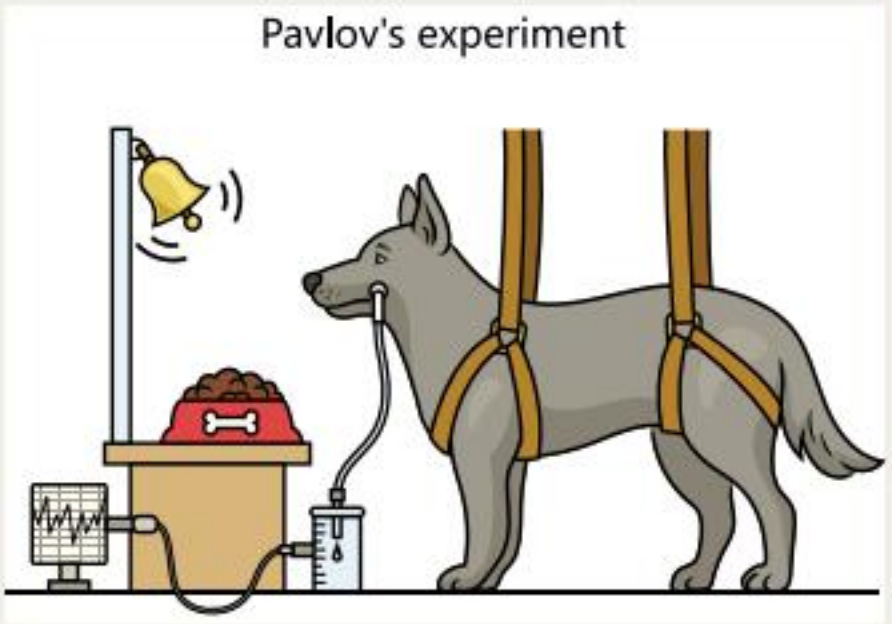
In the *Queensland Animal Care and Protection Act* 2001, a legal duty of care for animals is specified and pain is defined as including distress, mental or physical suffering to an animal.

Three basic principles known as the 3Rs are used globally in guidelines for animal-based research:

1 *replacement* of animals with non-living models
2 *reduction* in the use of animals
3 *refinement* of animal use practices.

Apart from medical, veterinary and scientific research, animal test subjects are widely used in agriculture, education, wildlife research and the pharmaceutical industry (Figure 10.3.2). The Australian Government's National Health and Medical Research Council (NHMRC) has specific guidelines, including '*A guide to the care and use of Australian native mammals in research and teaching (2014)*' with a long list of animals from echidna to quoll.

While animal-based research has its critics, others claim that total elimination of animal testing would significantly set back the development of essential medical devices, medicines and treatment for humans.



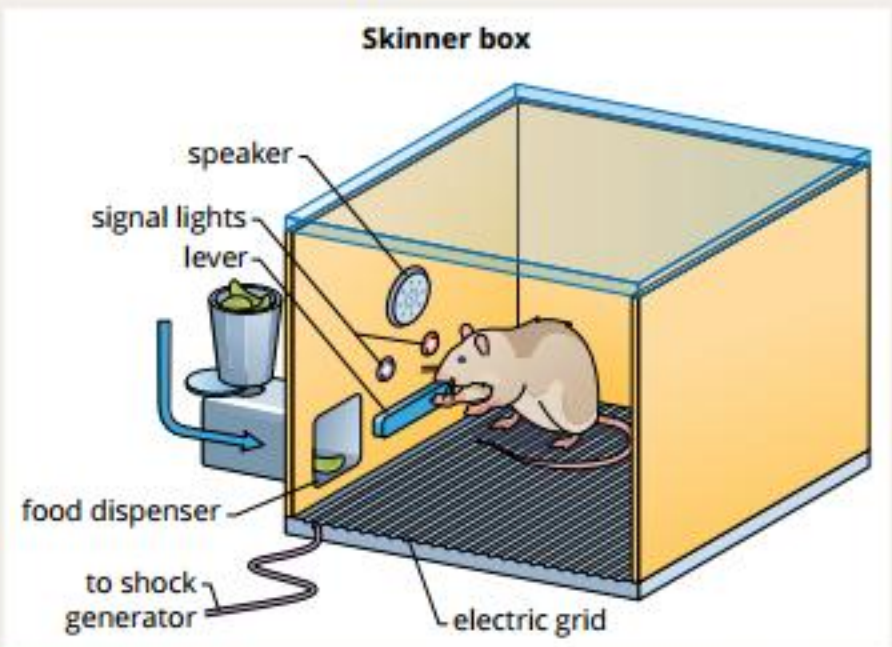


FIGURE 10.3.1 Two famous investigations of animal behaviour are (a) Pavlov's experiment that conditioned dogs to expect food when a bell rang and (b) Skinner's experiment in which a rat confined in a box would learn to press the lever to get a food reward.

FIGURE 10.3.2 A scientist uses humane practices for laboratory research with an animal.

KEY QUESTIONS

Forming and finding

1 Explain the concept of animals as sentient beings.

2 Summarise the contributions, outlined in this article, of animal-based research to scientific understanding of the human body.

3 To protect the welfare of animals used for research:
- **a** Is it necessary to have legal recognition of sentience?
- **b** How has recognition of animal sentience changed the view on using animals in research?

Analysing

4 Develop your understanding of the 3Rs by adding examples of how each could be put into practice in a school environment.

5 Discuss points for and against the continuation of animal-based scientific research. Support your discussion with examples on both sides of the debate.

Kidneys and healthy bodies

The kidneys are vital organs that filter toxic nitrogenous waste from the human circulatory system and maintain the water–salt ratio, thus preserving homeostatic balance in the body. Cleansing of the blood by the kidneys must be continuous for good health. The liver is also involved in processing nitrogenous waste from amino acids and nucleic acids into urea, ready to pass to the kidneys for excretion.

Kidney disorders lead to other health problems and can eventually be fatal. There are multiple causes for kidney problems. High-risk factors include hypertension (high blood pressure), diabetes types 1 and 2, smoking, obesity and a family history of kidney failure. Excessive alcohol consumption damages the liver and will, in turn, affect the kidney's ability to function.

The study of nephrology is a specialty area of medicine, and the structure and function of the renal system are well understood. Many hospitals have renal clinics to cater for patients with kidney failure, usually by dialysis treatment. Despite this, kidney disease is sometimes called a silent disease because there can be up to 90% loss of kidney function before any symptoms are recognised. Symptoms develop slowly and are not specific to kidney disorders. They include fatigue, high blood pressure, blood in the urine, pain in the lower back, nausea, loss of appetite, abnormal heart rhythm, urinating more or less frequently, itchy skin, puffy eyes and swollen ankles due to water–salt imbalance. Taken together, this list of symptoms appears alarming and easily detected. However, patients may exhibit minor or no symptoms initially and simply overlook them, or attribute them to other health issues such as diabetes, eating disorders, prostate problems or weak bladder muscles.

Because of the close connection between the liver, kidneys and circulatory system, doctors test both blood and urine samples to check for kidney and liver health. The tests are relatively simple, developed from basic biochemistry.

- A blood serum sample is used to measure a waste product called creatinine. The creatinine level is used in a formula with age and gender to calculate eGFR (estimated glomerular filtration rate). The eGFR is reported as mL/min/1.73 m^2 (1.73 m^2 is a reference value for body surface area). The eGFR is normal at ≥90 mL/min/m^2 or shows some reduced kidney function at 60–89 mL/min/m^2. Results consistently below 60 mL/min/1.73 m^2 for more than three months indicate chronic kidney disease. Results for eGFR are not valid for people under 18 years old or during pregnancy.
- Another diagnostic test, used with eGFR, is for the urea level in the blood. This blood urea nitrogen (BUN) test is less accurate on its own as it is affected by diet, hydration level and some medications.
- A urine sample is checked for any blood and to measure albumin. Albumin is a protein molecule that is too large to pass into Bowman's capsule in healthy kidney nephrons. In damaged nephrons, for example from an injury to the kidney area, some albumin may pass through the filter and into the urine.
- Other checks include monitoring blood pressure, testing for diabetes and imaging tests.

Based on the test results and a range of other considerations, a medical diagnosis of kidney disease may be made.

- Acute kidney disease (AKD) is where the kidneys recover normal function within three months. It may occur due to infection, blood loss, a heart attack or a sports injury. This is a reason why kidney punches are illegal in boxing, kickboxing and MMA.
- Chronic kidney disease (CKD) is a loss of healthy kidney function for more than three months. Kidneys at this stage cannot repair themselves. Careful management will extend life but eventually the kidney(s) will fail, and the only options are dialysis treatment (see Figure 10.4.1) or a kidney transplant.
- If one of their two kidneys is healthy, a person can still live a normal active life. A patient who receives a kidney transplant usually has one new kidney placed in their lower abdomen at the front of the body, with their diseased kidneys left in place (Figure 10.4.2).

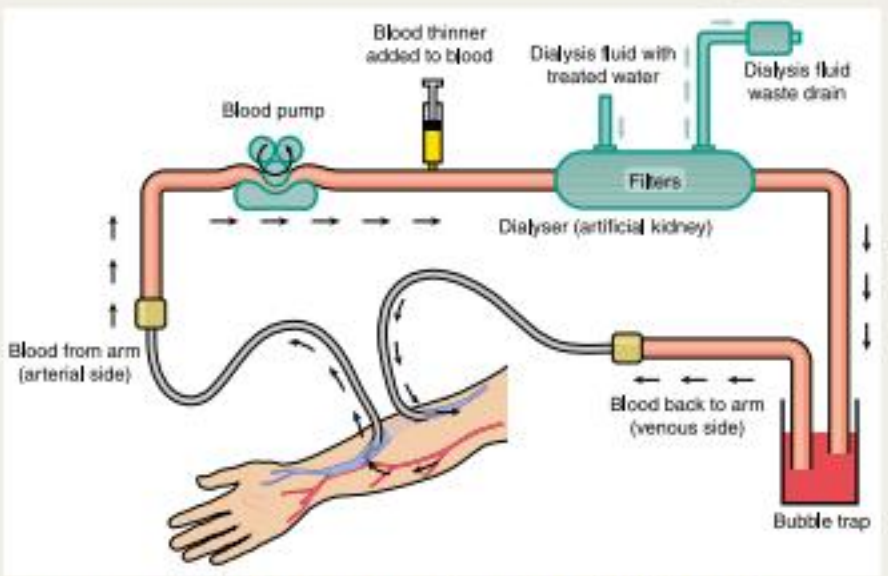


FIGURE 10.4.1 Dialysis treatment is used to artificially purify the blood of a person with kidney failure.

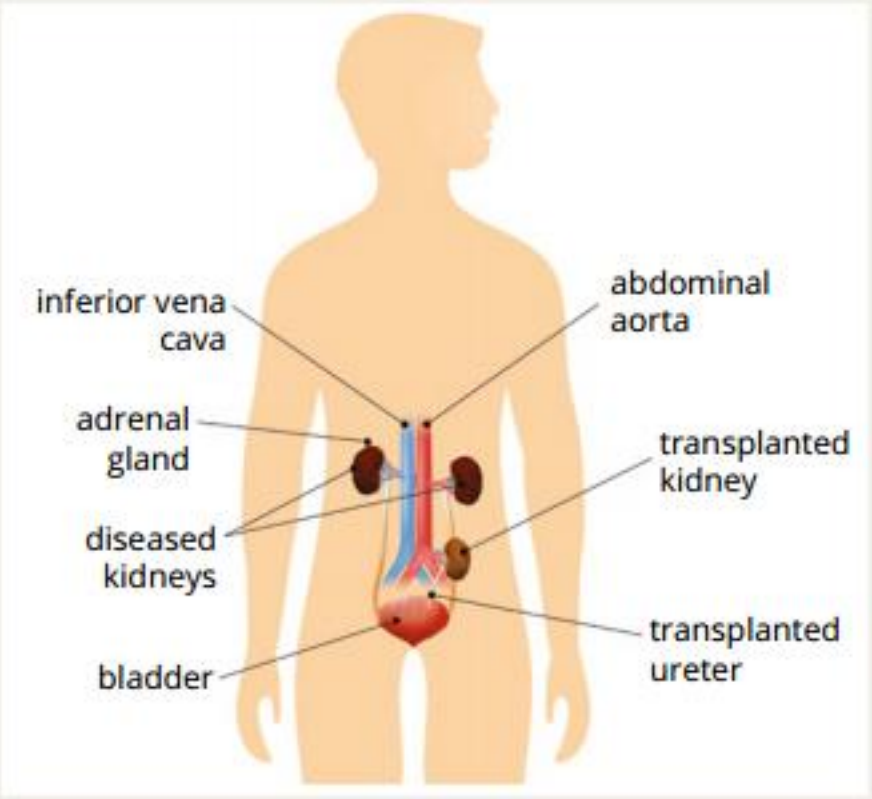


FIGURE 10.4.2 A kidney transplant involves a single healthy kidney with a ureter being transplanted and connected to the patient's blood vessels and bladder.

KEY QUESTIONS

Forming and finding

1 Recognise the relationship between the terms 'renal' and 'nephrology'.

2 In your own words, explain the interaction between the renal and hepatic (liver) organs with the circulatory system of the body.

3 Explain the difference between a BUN test and eGFR.

4 Visit the website of Kidney Health Australia.

- **a** Select a page on the Kidney Health Australia website and write the APA 7 reference for this page.
- **b** Use the 'About Us' page to judge if the website is a credible source.
- **c** List three useful facts about kidney health that you learnt from this website.
- **d** Write a paragraph about the Big Red Kidney Bus to convince a dialysis patient to plan a holiday.

Analysing

5 A normal eGFR of 100 mL/min is taken as having 100% kidney function.

- **a** If a person has a test result of eGFR equal to 15 mL/min, what is their percentage kidney function?
- **b** What is their likely diagnosis and future health outlook?

6 Predict whether a doctor would use dialysis treatment or a kidney transplant for a patient newly diagnosed with chronic kidney failure.

Cool clothing

As endothermic organisms, humans metabolise 70–100 W (watts) of heat when resting. However, under physical stress or exertion, adults can generate >1000 W of heat energy. If metabolic heat is not dissipated in time, it results in an increase in body temperature. When body temperature exceeds a certain range (hyperthermia), dangerous heat-related diseases such as heat stroke and dehydration may occur. This becomes an issue when the person wearing PPE is fighting a fire, working near furnaces or in toxic waste disposal, or perhaps is on a spacewalk around the International Space Station attending to repairs. The excessive heat load being generated cannot be dissipated by the normal means of human thermoregulation. Instead, the heat is retained by the wearer's protective clothing.

Since 1959, research has been conducted to design and manufacture protective clothing that has built-in cooling systems. A liquid cooling system (based on the efficiency of the human vascular system) can be used to carry body heat away from the skin surface of the wearer and dissipate it into the surrounding environment. The design of Microclimate Liquid Cooling Systems ($_{LQ}$MCSs) has been a priority for bioengineers and different versions of $_{LQ}$MCSs are commonly used today (Figure 10.5.1).

FIGURE 10.5.1 Current $_{LQ}$MCSs such as this spacesuit, are bulky and not as sophisticated as the human thermoregulatory system. Research is ongoing to make the suits more thermoregulatory and ergonomically efficient.

However, their efficiency, manoeuvrability and wearability vary greatly. In a 2006 research paper titled *Design and Control Optimization of Microclimate Liquid Cooling Systems Underneath Protective Clothing*, the authors, A.D. Flouris and C.C. Cheung, suggested the following areas for future research and development of $_{LQ}$MCSs:

- Replace polyvinyl chloride as the transport tube because it is a natural heat insulator.
- Because of manufacturing difficulties, garments are all of a generic size, however, one size never fits all.
- Ensure that tubing is evenly spread throughout the whole garment.
- There is no integrated 'command control' centre. More research is required in order to achieve an optimum automatic control design for $_{LQ}$MCSs.

More recent research has focused on the use of phase change material (PCM) and integrating it with cooling clothing. PCM provides both heating and cooling because it can either release or absorb energy when it changes state between solid and liquid. PCMs have many applications in buildings and refrigeration systems to passively regulate ambient temperature. The most commonly used PCMs are salt hydrates, fatty acids and esters, and various paraffins. Engineers must design suitable containers with regard to fire and personal safety for PCMs, depending on their application, such as numerous small, sealed pockets in a vest for a person to wear.

One investigation aimed to use PCM to improve the thermal comfort of healthcare workers during the COVID–19 pandemic while they were wearing PPE (personal protective equipment). Wearing PCM was found to reduce head and facial discomfort by 25% and 41%, respectively, under a 26°C thermal environment. An additional load of PPE in high-temperature and high-humidity working environments threatens the safety of health workers. This was an important lesson learned during the Ebola outbreak when working in hot parts of Africa restricted disease control methods. Hence, the value of research efforts to develop thermal regulatory clothing.

Workers and researchers in sub-zero environments traditionally wear layers of wool, fleece and waterproof clothing and live in insulated buildings to avoid hypothermia. The new technologies offer potential applications for them in the future as research and development of PCMs advances.

KEY QUESTIONS

Forming and finding

1 Distinguish between hyperthermia and hypothermia.

2 Define the acronyms used in this article: $_{LQ}$MCSs and PCMs.

3 Assess why use of PCMs is valuable in conjunction with cooling clothing.

4 Compare the similarities of the human thermoregulatory system with $_{LQ}$MCSs and contrast their differences.

Analysing

5 Using your knowledge of human thermoregulation, determine what components need to be present when designing an 'integrated command centre' for an $_{LQ}$MCS.

Enzymes and homeostasis

Cells use enzymes to regulate metabolism (all the body's biochemical reactions). When cells produce too much or too little of particular substances, or cannot break down substances, the whole organism is affected.

Specific enzymes bind to their matching substrates to catalyse particular chemical reactions. This gives the cell control over metabolic activity. All enzymes are protein molecules with a 3D shape that is critical to enzyme activity. You will recall studying enzymes in Module 3.2 (Enzymes and biochemical pathways) and that high temperatures denature (disrupt) the enzyme's active site. For complex organisms, regulation of the internal environment (known as homeostasis) is critical for maintaining optimal conditions for enzyme activity.

Tolerance ranges of enzymes

Most enzymes have a narrow range of pH at which they function best. Examples are those in the lysosome described below, or the extracellular enzymes of the digestive system that you studied in Chapter 5 (Organ systems). For example, pepsin is a digestive enzyme with an optimum pH of 2.5 and a working range of pH 1–4.

Enzymes are also sensitive to temperature with some tolerance variability. The endothermic mammals are equipped for internal thermoregulation and also use behavioural strategies to maintain the best temperature for enzyme-controlled metabolism. Their enzymes will continue to work over a limited temperature range outside the optimum, but at a slower rate.

Humans maintain a core body temperature in the narrow range of 37.0°C ± 0.5°C. Outside this range, enzyme activity and metabolism are slower, tending to zero at the extremes of the tolerance range. Other vertebrates cope with wider ranges of temperature using various adaptations, including enzymes that are less temperature or pH sensitive (Figure 10.6.1).

The enzymes of each species, like their other adaptations, have evolved over time to enable the species to live successfully in their natural environment. Some examples are:

- Sloths: the three-fingered sloth (*Bradypus variegatus*) of Central America is known for its low metabolic rate and unusual daily variation up to 10°C in core temperature. Sloths have a low-calorie diet of leaves full of toxins, with long periods of microbial digestion, so their energy is limited. Studies have shown them to continue eating and depress their metabolic rate at high ambient temperatures without becoming torpid (inactive). It is postulated that microbial fermentation in their gut increases in warmer temperatures without any extra metabolic effort from the sloth (Figure 10.6.2a).
- Sooty grunter (*Hephaestus fuliginosus*): a species of fish in northern Queensland and the Northern Territory that inhabits large freshwater streams and sometimes hot springs. Sooty grunters tolerate acidic conditions to a pH of 4.0 and water temperatures of 12–34°C. The resilience of sooty grunters indicates unusually wide tolerance ranges for their enzymes (Figure 10.6.2b).
- Antarctic invertebrates: British studies of three Antarctic marine species – a bivalve mollusc, a limpet and a scallop — showed they are more temperature sensitive than similar species in warmer waters. They could survive a few days in water temperatures up to 10°C but lost their ability to swim or burrow, and their tissues swapped to anaerobic cellular respiration.

Cellular compartmentalisation

Cellular compartmentalisation in eukaryotic cells creates a micro-environment with optimal conditions for enzymes. Within each organelle, different pH and cofactor chemicals support particular enzymes. An example inside animal cells is the lysosome, a specialised sac containing about 40 types of enzymes, known as the hydrolytic enzyme mixture, that breaks down and recycles aged or defective biomolecules. These enzymes have optimal pH 5, whereas most other cellular enzymes have optimal pH 7. Hence, it is important to isolate lysosomal enzymes within the organelle, both to promote hydrolase function and to protect other cell contents.

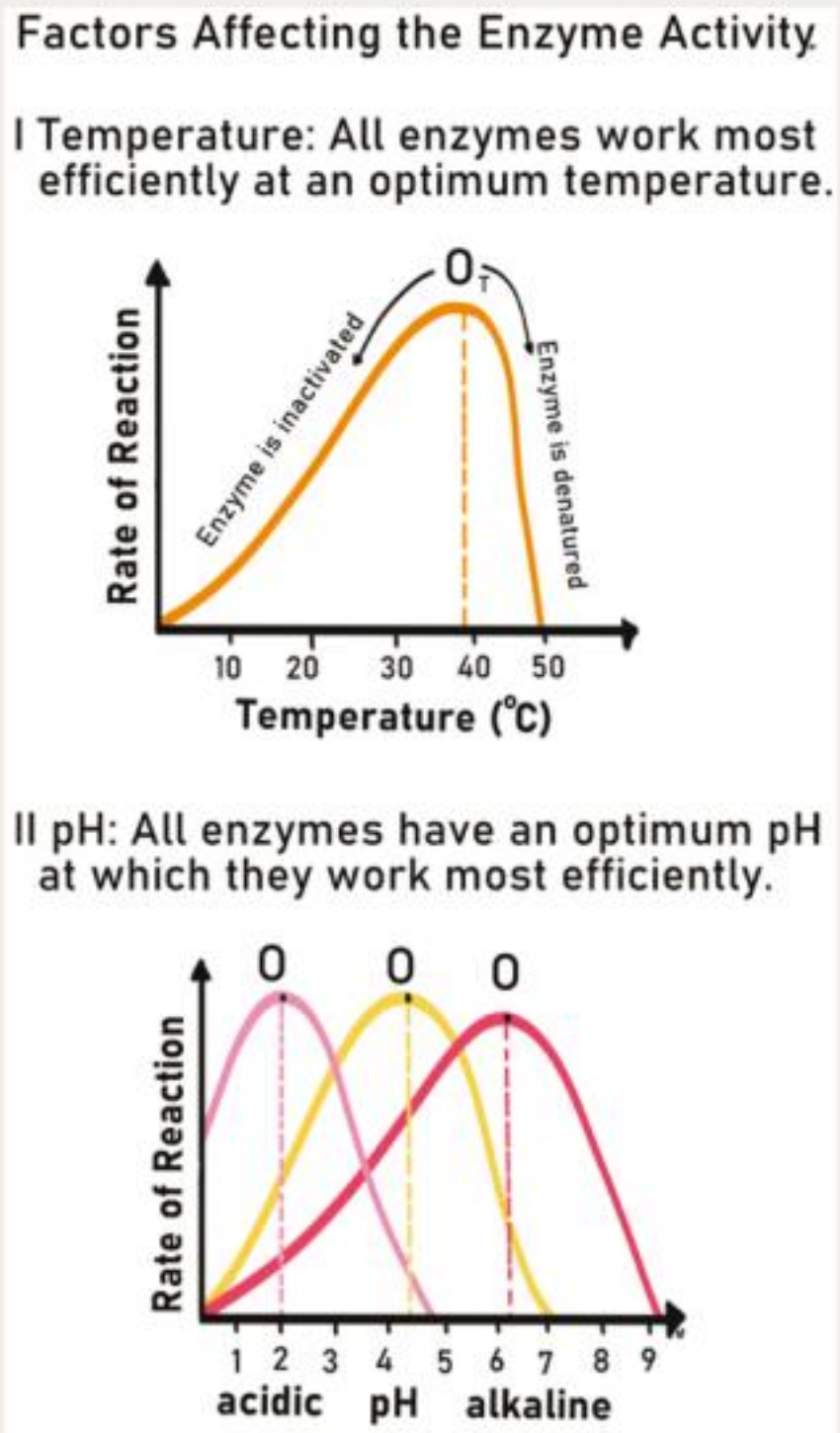

Factors Affecting the Enzyme Activity
I Temperature: All enzymes work most efficiently at an optimum temperature.
Rate of Reaction
Enzyme is inactivated
Enzyme is denatured
10 20 30 40 50
Temperature (°C)
II pH: All enzymes have an optimum pH at which they work most efficiently.
Rate of Reaction
1 2 3 4 5 6 7 8 9
acidic pH alkaline

FIGURE 10.6.1 Temperature and pH both affect the activity of an enzyme.

FIGURE 10.6.2 The enzymes of different species function within different pH and temperature ranges. (a) The three-toed sloth (*Bradypus. variegatus*) has an unusually wide tolerance range of core temperature for a mammal. (b) The sooty grunter (*Hephaestus. fuliginosus*) has unique metabolic enzymes that function over wide pH and temperature ranges.

KEY QUESTIONS

Forming and finding

1 Identify two factors that can influence the activity of an enzyme.

2 Define the term 'tolerance range', using a specific example to support your answer.

3 Use secondary sources to find two more examples of organisms with unusual enzymatic tolerance ranges.

Analysing

4 Assess the advantages and disadvantages for complex organisms of regulating their internal environment.

5 The Australian koala, a marsupial, has a similar lifestyle to the three-toed sloth. Conduct research to compare the two animals.

Traditional medicines of First Nations Australians

First Nations peoples around the world use traditional knowledge of medical remedies from the natural resources at hand for their survival. With colonisation followed by globalisation, western medical practices tended to overwhelm the use of natural remedies. Now a new appreciation of traditional medicines has emerged as they are estimated to be used by 60% of the world's population.

As one of the world's oldest surviving cultures, First Nations peoples in Australia have a profound connection with Country and a deep knowledge of native flora and fauna. Traditionally, a healthy diet from bush tucker was the most important way to maintain their health. If people became sick or were injured, they used plants for healing and developed methods to use them safely.

Many of these traditional medicines and methods of healing are still used today. These methods include crushing, heating and applying plant material to the skin, boiling plant parts and inhaling the vapour or drinking the extract. Raw plant materials can also be applied, such as sap from a bloodwood tree, which acts as an antiseptic and can be smeared directly on cuts and sores. Soil sediments are also used as remedies with certain types used to prevent and treat sunburn, to cover wounds to prevent infection, or small amounts of clay swallowed to deactivate gastrointestinal toxins. Bush remedies were, and still are, accompanied by spiritual healing practices. Some grasses and barks are important in smoking and healing ceremonies.

The effectiveness of many medicinal plants has been supported by thousands of years of use by First Nations peoples in Australia and more recently, by scientific research. These medicinal plants include:

- Quandong (*Santalum acuminatum*), also known as wild peach, has fruit high in vitamin C and is a source of many minerals. The kernels are ground into an antibacterial paste to treat skin conditions and toothache.
- Maroon bush (*Scaevola spinescens*) (Figure 10.7.1) grows in most semi-arid parts of Australia and has traditionally been used for heart, kidney and intestinal ailments. Research has identified that maroon bush has strong antibacterial and antiviral properties. The properties are so potent that it is being investigated for use against bacteria and fungi that are resistant to existing antibiotics and antifungals. It is also being investigated for its potential as a broad-spectrum antiviral against RNA viruses that infect humans (e.g. coronavirus group: COVID–19, SARS and influenza; hepatitis C, measles).
- Emu bush leaves (*Eremophila sp.*) (Figure 10.7.2) are traditionally used to cleanse surface wounds or gargled to treat throat infection. The vapours are used to inhibit bacterial and fungal pathogens. Modern scientific research has discovered that the plant's properties are equal in strength to antibacterial agents such as the penicillins.
- Tea tree oil *(Melaleuca alternifolia)* (Figure 10.7.3) has achieved global use as a household antiseptic and to treat fungal infections. It was commonly used by the Bundjalung people from the NSW north coast where it originated. Demand is such that it is now widely grown in plantations.
- Eucalyptus oil can be extracted from the leaves of the many eucalypt species that are found across Australia. First Nations peoples use it to treat body pains, fevers and chills. Eucalyptus oil is used commercially as an antiseptic in mouthwash, throat lozenges and cough suppressants.
- Witchetty grubs *(Endoxyla leucomochla)* are enjoyed as a healthy natural food source in Central Australia. They are also used to treat burns by crushing them into a paste that is applied to the damaged skin. The grubs often feature in artworks from Central Australia.

There is still much to learn about contemporary applications for traditional medicines. As this knowledge develops, it is important to respect and protect the cultural and intellectual property rights of First Nations peoples, who are the rightful custodians of this knowledge.

FIGURE 10.7.1 The maroon bush (*Scaevola spinescens*) has highly potent antiseptic properties that are being investigated for commercial use.

FIGURE 10.7.2 Emu bush (*Eremophila sp.*) has been used traditionally in bush medicine, and now in modern research, for its strong antibacterial properties.

FIGURE 10.7.3 Tea tree (*Melaleuca alternifolia*), native to coastal northern NSW, is used in traditional bush medicine and now in many commercial products.

KEY QUESTIONS

Forming and finding

1 Identify two ways that plants can be administered to heal a patient in traditional bush medicine.

2 Create a table to summarise four traditional medicines noted in this article.

3 Make a list of any natural remedies used by your family or friends. Identify any that may have started as traditional knowledge.

Analysing

4 First Nations peoples had no written language prior to colonisation.
   - **a** How has their medical knowledge been passed down through generations?
   - **b** Suggest how they have developed their knowledge without scientific laboratories to analyse and test substances.
   - **c** How have modern pharmaceutical companies acquired and applied some of this traditional knowledge to their products?

5 Discuss the issue of who 'owns' traditional medicines taken from the bush and what financial protocols are involved.

Scientific advancement through collaboration

Just as every film rolls through a long list of credits at the end acknowledging those involved, even the solo rock stars of the science world have a support team, brainstorm with other scientists and are inspired by discoveries of earlier scientists. The complexity of research, often using expensive specialist technologies, usually requires a team of experts. It is rare to see a scientific paper written by a single author.

Mostly gone is the 'little science' of a single investigator–inventor like Robert Hooke in the 1660s making his own microscope and slide preparations of cork cells. Scientific advancement now is more likely to be 'big science' requiring teams of people, processing of data and use of applied science to develop new technologies. 'Interdisciplinary', 'interdependent', 'international', along with 'collaboration' and 'communication', are key descriptors for much of modern scientific research.

While we can credit Rene Laennec as the solo inventor of the stethoscope in France in 1816, the path to developing something like the modern kidney dialysis machine was more complex. The technology of dialysers took more than a century to develop, as did scientific understanding of the transport of substances across membranes. Scientists finally learned how to quantify the dialysis process and reliably access a patient's vascular system with an arteriovenous (AV) fistula. Now the latest generation of dialysis equipment can be used at home with a computer-controlled machine and online technology.

From the eradication of smallpox to the management of Ebola and the COVID–19 pandemic, international collaboration in health science has been a major success. It has led to breakthroughs and advances at a speed unattainable for countries working alone. For example, in 2019–2020 a team of engineers, doctors and computer scientists at Massachusetts Institute of Technology in the USA worked on a low-cost ventilator for patients hospitalised with COVID–19. At the same time, its open-source design was used by a group of Indian robotic engineers to build ventilators to ease their country's shortage of the equipment.

Open-source models

Open-source design refers to publicly shared design information. This includes access to both software and hardware. The process is generally facilitated on the internet, often without financial compensation, and may lead to collaborations that merge basic and applied sciences. There are also specialist websites that link researchers with others in the same field or help them find volunteers to process data. Citizen science projects have proliferated as internet access expanded and smart technologies became available to non-specialists of all ages. The Australian Citizen Science Association works in partnership with the Atlas of Living Australia and has a Project Finder site to provide opportunities for volunteers.

International collaboration for the ISS

An outstanding example of scientific collaboration is the International Space Station (ISS), constructed and controlled jointly by agencies from the USA, Russia, Europe, Canada and Japan, and now with some private aerospace industries involved. This level of space technology would be out of reach for any single agency on its own. In June 2024, the dedicated NASA website noted that the ISS had been 25 years in orbit, had 270 visits from astronauts and could be described as:

> The International Space Station Program brings together international flight crews, multiple launch vehicles, globally distributed launch and flight operations, training, engineering, and development facilities, communications networks, and the international scientific research community.

The ISS has evolved into a state-of-the-art scientific laboratory, which will continue its orbit until at least 2030. Much of the research is being conducted in the areas of biology and biotechnology (Figure 10.8.1). For example, research using the space station's PCR machine and portable DNA sequencer (MinION) to study DNA in real time onboard is developing our understanding of epigenetics–a research topic not even conceived of when the ISS was launched.

FIGURE 10.8.1 A NASA astronaut at work on the ISS in 2020. This project is being conducted inside the Life Sciences Glovebox in Japan's laboratory module, to study the effects of space on heart tissue.

KEY QUESTIONS

Forming and finding

1 Distinguish between 'applied science' and 'science', providing an example of each.

2 In the second paragraph, there are five key words used for the nature of modern scientific research: interdisciplinary, interdependent, international, collaboration, communication. Choose one of the scientific developments described in this article (such as the ISS) and write your own outline of the project incorporating these five words.

3 Infer at least three reasons why the nature of scientific research has changed over time.

4 It took more than 100 years for reliable kidney dialysis to become available. Read the third paragraph again, then discuss if you think the equipment would have been developed faster in the 21st century.

Analysing

5 Predict what may have happened if there had not been international collaboration during the COVID–19 pandemic.

6 Select a citizen science project and explain how you would go about contributing to it.

Protecting our borders—Australian biosecurity

Infectious diseases cause significant illness and loss of life in the global human population. Over the ages, bubonic plague, smallpox, malaria, Ebola and recently the COVID–19 pandemic are some of the most devastating examples. There are more, including the emergence of new zoonotic diseases as the world's human population expands and moves into closer contact with animals. A zoonosis is an infectious disease that jumps from non-human animals to humans, such as the rabies virus spread from an infected dog to a human, or the Hendra virus spread from a fruit bat to a horse to a human.

Rapid and increasingly frequent movements of people between countries and large cities across the globe facilitate the spread of diseases. Australia has a strong emphasis on biosecurity to prevent, respond to and recover rapidly from pests and diseases. This helps secure our agricultural industries, biodiversity and human health.

New diseases can have severe effects when the human population has no previous resistance to them. For example, the 2009 H1N1 swine flu virus has been traced to pig farms in a small area of central Mexico. The parent virus is believed to have existed for more than 10 years in pigs before a viral strain developed with the capacity to infect humans. As a new strain of influenza virus, humans had little immunity to it. This virulence highlighted the need for worldwide surveillance and data collection for both animal and human viruses on local, regional and global scales. Strict quarantine measures are used to isolate and destroy plants and animals infected with a disease to contain further outbreaks.

Some examples of foreign diseases that have entered Australia are:

- Panama disease, a fungal infection in banana plants, spread to North Queensland in 2015 from other countries. Case Study 7.2.2 (page 287) describes the dangers of it infecting nearby plantations.
- Outbreaks of bird (avian) flu reoccur in Australia from time to time, causing significant mortality to domestic poultry and wild birds. Each state has a plan to detect, manage and eliminate outbreaks.
- Foot-and-mouth disease infects farm livestock and is a continuing high-level biosecurity risk to the agriculture industry due to Australia's proximity to Southeast Asia, where it is endemic (Figure 10.9.1).
- *Varroa* mite is a serious parasite of the European honeybee but does not infect native bees.

  In 2022, the first mites were found in sentinel hives in the Port of Newcastle. Despite a rapid multi-agency response, initial efforts to eradicate the mite failed and it has become established in NSW. Beekeepers are now reporting any mite detection and being educated in how to manage healthy beehives for honey production and to maintain pollination of crops despite the presence of *Varroa* mites (Figure 10.9.2).

Biosecurity measures at airports, border stations and seaports include entry and exit screening, searching luggage, remote body temperature-sensing, health-alert notifications, collection of passenger/cargo information, public health reporting, and physically examining sick people and animals (Figure 10.9.3). Inspecting goods in private or commercial travel is also a quarantine practice, with contaminated animals or goods inspected, disinfected, fumigated, isolated or destroyed. Australia has very strict quarantine laws, enforced by the Australian Quarantine and Inspection Service (AQIS), which prohibit the entry of people, animals or goods that are considered a risk, as well as enforced isolation of anyone infected. State borders in Australia, and some local areas, also have quarantine, such as banning the movement of fruit across borders to prevent the spread of fruit fly.

In May 2024, Queensland launched a new five-year biosecurity strategy for combating plant and animal pests and diseases. It includes a focus on community awareness and that biosecurity is everyone's responsibility.

FIGURE 10.9.1 Foot-and-mouth disease in livestock spreads rapidly and is carried on footwear.

FIGURE 10.9.2 Honeybee with *Varroa* mite attached

FIGURE 10.9.3 Quarantine officers and sniffer dogs help inspect and screen humans, animals and goods at airports and other borders.

KEY QUESTIONS

Forming and finding

1 Define the two terms in this article that may be new to you: 'zoonotic disease' and 'sentinel hive'.

2 Has the *Varroa* mite spread to beehives in Queensland?
   - **a** Use credible sources to find out.
   - **b** Explain why you believe your source(s) to be reliable.
   - **c** Cite your source(s).

3 List the biosecurity checks used for entry into Australia in the order that you think are most to least effective.

Analysing

4 Discuss whether it would be possible to prevent any new diseases from entering Australia.

5 Describe in a short paragraph any personal experience you have had with biosecurity measures.

Vaccination programs

Vaccination is used to prevent infectious diseases caused by pathogens and transmitted between humans. For non-infectious diseases such as cancers, diabetes or heart and kidney diseases, there are currently no vaccines to directly prevent them. However, there are some vaccines that can indirectly reduce the risk of developing certain non-infectious diseases, such as the HPV (human papillomavirus) vaccine that reduces the risk of infection with a virus (HPV) that is linked to cervical cancer.

Vaccination programs play an important role in preventing the spread of infectious disease and keeping populations healthy. Data from epidemiological studies underpins all mass vaccination programs in Australia.

Pandemic, epidemic and endemic disease

The WHO (World Health Organization), a United Nations agency, defines pandemics, epidemics and endemic diseases based on each disease's rate of spread. The difference between an epidemic and a pandemic is not the severity of the disease, but the degree to which it has spread. An epidemic is a sudden increase in the incidence of a disease in a localised region. A pandemic is an epidemic that has spread over a larger geographic area and can potentially affect people across multiple countries or continents. An endemic disease is consistently present with a predictable and low rate of infection. Common influenza is classified as an endemic disease, however, there can be outbreaks that lead to a flu epidemic or even a pandemic such as H1N1 (swine flu) in 2019. With the COVID–19 pandemic, restrictions, vaccinations and treatments have now reduced this disease to endemic status in Australia.

The disease outbreak models

The nature of an infectious disease, how the infection might spread and the number of people likely to be infected are essential factors to consider when planning intervention measures such as a vaccination program. Mass vaccination is more likely to succeed for a human-to-human infection where recovery confers active immunity, such as with measles, mumps and rubella. For some diseases, vaccination in early childhood is most effective, e.g. varicella (chicken pox) at 18 months of age, which then prevents reactivation as shingles in older adults. A current shingles vaccination program for adults over 65 years of age who are at risk should reduce as the early-vaccinated generations come through.

The strategy for a disease that is transmitted from animal to human is early detection and control rather than eradication. This is because transmissible infection will remain in the animals, such as bird (avian) flu in wild birds or brucellosis in cattle.

Eradication was possible in the case of smallpox, a disfiguring and deadly virus infection. On 8 May 1980, the WHO officially announced that smallpox had been eradicated in October 1979. Since then, there have been no vaccinations against smallpox. Australian virologist, Frank Fenner (1914–2010) (Figure 10.10.1), made that momentous announcement. Fenner helped establish that monkey populations were not acting as a reservoir for smallpox, demonstrating that eradication was possible. He then led the global campaign of intensive intervention with the cooperation of governments across the world. They used a 'ring vaccination' model around each outbreak and tracked down every reported infection no matter how remote. This technique led to successful eradication. Smallpox is the only disease to date that has been successfully eradicated, though polio is now close to elimination. Factors that contributed to smallpox eradication included lasting immunity in those who recovered, as well as the smallpox virus only infecting humans.

Using mathematical modelling to assess early information about an epidemic can predict its future course weeks in advance, helping authorities to plan effective responses, proactively as well as reactively. Measles is one of the earliest pathogens to be modelled, dating back to the late 1800s. Most recently, the 'SEIR' model was applied in Australia to help manage the COVID–19 pandemic:

- S – all individuals are assumed to be susceptible to infection at the outset of the outbreak
- E – exposed to the virus when they come into contact with I (see below)
- I – an infected person (as exposed individuals become infected and infectious)
- R – once recovered, individuals are assumed to be fully resistant to reinfection, at least for a time.

Researchers can refine the model by including data from actual outbreaks as they become available.

Australia's national free vaccination program has been carefully developed from epidemiological data to be most effective. Records are maintained of notifiable diseases and the Australian Immunisation Register (AIR) records all vaccines given to people in Australia. This includes COVID–19 vaccines, vaccines given under the National Immunisation Program and vaccines given privately, such as for seasonal influenza or travel.

FIGURE 10.10.1 Professor Frank Fenner led the global campaign for WHO to eradicate smallpox. This was accomplished by ring vaccination of populations around each outbreak, rather than a mass vaccination program.

KEY QUESTIONS

Forming and finding

1. Distinguish between the terms 'endemic', 'epidemic' and 'pandemic' when applied to infectious diseases. Use an example of each term to support your answer.
2. List at least three important factors that make a mass vaccination program successful.
3. Explain why it is important to know whether a disease is only transmitted human to human, or if an animal to human transmission is involved.
4. Use a flow diagram to summarise the SEIR model for a disease outbreak.

Analysing

5. Currently, smallpox is the only disease that has been eradicated. Efforts to eradicate polio are managed by GPEI. Find a scientifically credible resource for polio information.
   - **a** Identify which countries still have polio infections.
   - **b** Do your own research to recognise three points of difference with the smallpox campaign.
   - **c** Explain what needs to be done to completely eradicate polio.
   - **d** Cite your source(s) correctly.

Glossary

A

abscisic acid (ABA) A growth-inhibiting plant hormone associated with leaf and fruit fall, as well as bud and seed dormancy.

absolute uncertainty The uncertainty of a measurement expressed in the same units as the measurement.

accuracy The quality of being true, correct or precise, representing the extent to which a measurement reflects the actual quantity being measured. In science, accuracy requires both obtaining a measurement close to the true value and providing an estimate of uncertainty. Achieving accurate results requires minimising systematic errors to ensure the measurement reliably captures the intended quantity.

action potential A reversal of the normal potential difference across a cell membrane, or between the inside and outside of a nerve fibre; allows the organ to carry out its function such as contraction of the heart.

activation energy The energy that is required to start a biochemical reaction.

active immunity Immunity that involves an individual's own adaptive immune response, through B and T lymphocytes.

active site The specific site of an enzyme that binds the substrate and where catalysis occurs.

active transport The movement of substances across membranes that requires the expenditure of energy; occurs through selective protein channels.

adaptation (1) An inherited character that increases the likelihood of survival and reproduction of an organism or species. (2) The process by which a species becomes well-suited to its lifestyle and environment.

adaptive immune response An immune response that is modified on each subsequent infection to become more effective and to provide long-term immunity to pathogens.

adenosine triphosphate (ATP) A molecule that provides energy for immediate use by a cell; produced during glycolysis and cellular respiration.

adherence factor Surface molecule on a pathogen that enables it to attach to host cells or tissues, facilitating infection.

adhesin A structure or molecule on the surface of bacteria, which allows them to attach to their prospective host cell.

adjuvant A chemical added to a vaccine that enhances the ability of the vaccine to induce an immune response.

adrenaline A hormone released from the adrenal glands responsible for sympathetic nervous system activation including increased heart rate, rapid breathing and vasodilation of pupils.

adult stem cell A stem cell that is present in some adult tissues. Adult stem cells can give rise to only a limited range of cells.

aeciospore A small, usually single-celled, asexual body generally containing two nuclei.

aerobic Requiring oxygen.

aerobic respiration Cellular respiration being undertaken in the presence of oxygen.

aestivation A period of prolonged torpor or dormancy of an insect, fish or amphibian during a hot or dry period.

afferent (sensory) neuron A neuron that communicates information from tissues and organs to the central nervous system.

agglutination The process in which antibodies bind to antigens on the surface of cells and form antigen–antibody complexes that clump together and activate phagocytes and the complement cascade.

agricultural pest An organism that is detrimental or a nuisance to agriculture, often causing disease.

air sac A lung compartment containing air.

aldosterone A hormone released from the adrenal glands that simultaneously regulates sodium and potassium levels by increasing potassium excretion into the urine and causing sodium reabsorption into the blood.

alkaloid An often toxic chemical produced by plants, including caffeine and nicotine, and which helps protect the plant from pathogens.

allele An alternative form of a gene.

allergen An antigen that elicits an allergic response.

alveoli (sing. alveolus) A small air sac in the lungs of mammals, located at the ends of the bronchioles. Alveoli have very thin cell membranes surrounded by a network of capillaries, which enables efficient gas exchange across their surface.

amino acid An organic compound containing an amino group ($-NH_2$) and a carboxyl group ($-COOH$) at opposite ends of the molecule. Linked amino acids form the peptide chains in protein molecules.

ammonia A compound (NH_3) that is the first nitrogenous waste to be formed from the breakdown of proteins. Ammonia is highly toxic and is excreted mainly by aquatic animals.

amylase An enzyme that breaks down starch molecules.

anabolic reaction Biochemical reaction that produces larger molecules from smaller substrates. Anabolic reactions are endergonic because they require energy to form bonds between molecules.

anaerobic respiration Cellular respiration being undertaken in the absence of oxygen.

anomaly A deviation from what is expected or considered normal.

antibacterial An agent or substance that works against bacteria.

antibiotic A substance, produced by a microorganism or synthesised, that inhibits the growth of a type of bacteria.

antibody A protein secreted by plasma cells that binds to a particular antigen and marks it for elimination; also known as immunoglobulin.

antidiuretic hormone (ADH) A hormone secreted by the pituitary gland that increases the permeability of the collecting tubule of the mammalian kidney to water. ADH increases the amount of water absorbed and so causes a smaller volume of more concentrated urine to be formed.

antigen A substance capable of stimulating an immune response.

antigen–antibody complex A specific chemical interaction between an antibody (immunoglobulin) molecule and an antigen molecule.

antigen-presenting cell (APC) A cell that uses MHC-II on its surface to present foreign antigens to helper T lymphocytes to elicit an adaptive immune response.

antigenic drift A slow change in the antigens on the surface of a pathogenic organism due to random mutations.

antigenic shift An abrupt change in the genetic code of a virus due to re-assortment of genes from different viral strains, resulting in significantly different antigens on the coat of the virus.

antiporter A protein carrier molecule found in the cell membrane that transports two different molecules across the membrane in opposite directions.

antiserum An aliquot (measured volume) of blood serum that contains specific antibodies against a specific antigen; is used to induce artificial passive immunity.

aorta The large artery that carries blood from the left ventricle of the heart to the body.

aquaporin A channel protein that facilitates the movement of water across the cell membrane.

archaea Unicellular prokaryotic organisms that belong to the domain Archaea. Archaea includes organisms that can live at high temperatures (thermophiles), in acidic environments (acidophiles) or very salty environments (halophiles). See also bacteria, prokaryote.

arteriole A small blood vessel that stems from an artery and leads to capillaries.

artery A blood vessel with thick, elastic walls, through which blood flows from the heart to the rest of the body.

artificial active immunity The immunity induced by injection of antibodies produced by another organism.

artificial passive immunity The immunity induced by administration of a serum containing antibodies made in another organism.

asymptomatic To be without symptoms, or not showing symptoms while infected and/or diseased.

atrium (pl. atria) A chamber of the heart that receives blood returning from the body or the lungs, before passing the blood into a ventricle. The left atrium receives oxygenated blood from the lungs, and the right ventricle receives deoxygenated blood from the rest of the body.

autonomic nervous system A subdivision of the nervous system that regulates the internal environment.

autotroph An organism that can produce its own food from inorganic materials, using light or chemical energy. Plants that photosynthesise are the most common autotrophs. All autotrophs are producers.

auxin A plant hormone that promotes the growth of new shoots, phototropic responses and the development of leaves and fruit.

axon The part of a neuron (nerve cell) that conducts an action potential away from the cell body towards the next neuron.

axon terminal Tip of a neuron's axon that releases neurotransmitters to communicate with other cells.

B

B lymphocyte (or B cell) A white blood cell that when stimulated produces large quantities of antibodies specific to a particular antigen. They are responsible for the humoral immune response.

bacteria (sing. bacterium) Unicellular prokaryotic organisms that belong to the domain Bacteria.

bacteriophage A virus that infects bacteria.

bar graph A graph that shows the measured value (or value of central tendency) of the dependent variable by the length of the horizontal bar.

baroreceptor A receptor that detects changes in blood pressure, which is an indication of the volume of blood.

basidiospore A haploid reproductive cell produced by meiosis that can divide and develop into a haploid fungus.

bias An inclination or prejudice towards something.

bile A secretion produced by the liver and stored in the gall bladder, from where it is released into the small intestine. Bile acts as an emulsifying agent, physically breaking up large fat droplets into smaller droplets to increase the surface area of food being digested.

biofilm A structured community of microorganisms embedded in a self-produced extracellular matrix of polysaccharides, proteins, and DNA. While bacteria are the most common components of biofilms, other microorganisms such as fungi, algae, and protozoa can also contribute to or form biofilms independently.

biogenesis The hypothesis that living matter arises only from other living matter.

bladder A muscular organ that receives urine from the kidneys and holds it before it is excreted through the urethra.

blastocyst The blastula stage in the development of a mammalian embryo; consists of an outer layer of cells that develop into the placenta, an inner mass of cells that develop into the embryo, and a fluid-filled cavity.

Bowman's capsule The region of a nephron into which filtered plasma flows from the glomerulus.

bronchi (sing. bronchus) An airway in the respiratory tract that conducts air into the lungs.

C

caecum An intestinal pouch at the junction of the small and large intestine. In some herbivores, such as koalas, it is very enlarged and acts as a fermentation chamber for the digestion of cellulose.

calibration The adjustment of an instrument with a standard scale of readings.

capillary A tiny blood vessel with a wall only one cell thick, across which exchange occurs between blood and tissues.

capsid The protein coat of a virus.

capsule A polysaccharide layer that coats the outside of the cell wall of some species of bacteria.

carbohydrate An organic compound consisting only of carbon, hydrogen and oxygen atoms, with the hydrogen and oxygen atoms in the same proportion as in water (2:1). Carbohydrates include sugars, starches and cellulose.

carbon dioxide (CO_2) Colourless, odourless gas that is used during photosynthesis and formed during cellular respiration.

carnivore An organism that feeds only on other animals.

carotid rete system A configuration of blood vessels from the carotid artery in the sinus cavity that cools the brain in some mammals.

carrier (1) An organism infected by a pathogen, usually without being affected by the pathogen, which can transmit the pathogen to another organism. (2) A heterozygous organism with an allele for a genetic disease which is not presented as the other allele is expressed.

carrier protein A protein that transports a specific substance across the cell membrane.

Casparian strip A water-resistant strip in the endodermis of roots that regulates the entry of water and solutes.

catabolic reaction Biochemical reaction in which there is a breakdown of macromolecules into smaller molecules. Catabolic reactions are exergonic because they release energy.

catalyst A substance that accelerates the rate of a chemical reaction by lowering the activation energy, without being used up in the reaction.

catecholamine An amine derivative of catechol that acts as a neurotransmitter or hormone (e.g. adrenaline and dopamine).

cell The smallest structural and organisational unit of which all living things are built.

cell body Part of a neuron (nerve cell) that contains the nucleus and other organelles.

cell compartmentalisation The characteristic of eukaryotic cells where the cell is divided into functional membrane-enclosed regions (organelles) with specialised functions.

cell differentiation The process by which a cell changes from one type to another. This is usually an unspecialised cell becoming a specialised cell.

cell-mediated immunity An immune response that is mediated by T lymphocytes.

cell membrane The plasma membrane at the boundary of every cell that acts as a selective barrier, thereby regulating the chemical composition of the cell.

cell potency The potential of a stem cell to differentiate into other cell types. The more cell types a stem cell can differentiate into, the greater its potency.

cell wall An external structure that surrounds the cell membrane for structural support and protection. Composed of cellulose (in plants) or peptidoglycan (in bacteria).

cellular respiration The energy-releasing processes that occur in cells. In particular, the aerobic stage in the complete breakdown of glucose to produce ATP, which occurs in mitochondria and produces 30–38 molecules of ATP per molecule of glucose.

cellulase Enzyme that catalyses the breakdown of cellulose.

cellulose A complex carbohydrate molecule consisting of a chain of many glucose molecules. It is the main component of plant cell walls. Its formula is $(C_6H_{10}O_5)_n$.

central nervous system (CNS) The brain and spinal cord in vertebrates.

channel protein A specialised protein that allows the transport of specific substances across a cell membrane.

chemical (HAZCHEM) code A system of codes and images that provides warnings of hazards about items to users.

chemical digestion The action of enzymes in breaking down complex compounds into simple compounds that can be used for metabolism.

chemokine A cytokine that attracts leukocytes to the site of an infection.

chemoreceptor A sensory receptor that detects and responds to specific chemical substances.

chlamydospore A resting spore with a thick cell wall that is produced asexually.

chloroplast A membrane-bound organelle involved in photosynthesis.

cholesterol A steroid found in cell membranes of animal tissues.

chromosome A structure made of DNA that encodes all of the information needed to make and operate a cell.

cilia (sing. cilium) A hair-like structure on the surface of some eukaryotic cells, consisting of a '9 + 2' arrangement of microtubules enclosed by an extension of the cell membrane. Cilia move with an oar-like motion and are usually shorter and more numerous than flagella.

claim An assertion made to influence decision-making or public debate. It represents a proposed explanation, interpretation or conclusion, and is subject to evaluation and scrutiny in scientific investigation.

clonal expansion The process of a particular B lymphocyte dividing rapidly to form many plasma cells that will produce specific antibodies.

clonal selection The theory that in a group of lymphocytes, a specific antigen will activate only the lymphocyte that has a receptor that specifically recognises it. This lymphocyte will then undergo clonal expansion.

closed circulatory system A circulatory system in which the fluid (blood) is confined to vessels and is kept separate from the interstitial fluid.

cluster A larger than expected number of cases of disease occurring in a particular locality, group of people, or period of time.

coefficient of determination A calculated value that indicates the ability to predict the dependent variable value from the independent value.

coenzyme A small organic molecule that combines with an enzyme and is necessary for its activity.

cofactor A metal ion or an organic molecule that is required for enzyme activity.

column graph A graph that shows the measured value (or value of central tendency) of the dependent variable by the height of the column.

communicable disease An infectious disease that can be transmitted from person to person within a community.

community immunity The phenomenon in which indirect protection from a pathogen is achieved through the immunity of a large proportion of the population, providing protection to non-immune or non-vaccinated individuals. Often achieved through vaccination and is also known as herd immunity.

competitive inhibition When an inhibitor has the same binding site as a substrate, they will compete for access to the enzyme.

complement protein A protein that can kill foreign cells by lysis. There are more than 30 different complement proteins, which are activated in response to antigen–antibody complexes, antigens and carbohydrates on the surface of some bacteria and parasites.

complement system A component of the innate immune response which complements, or enhances, the activity of antibodies and phagocytes, activates inflammation and immune clearance, and ruptures membranes of foreign cells.

concentration gradient The difference in concentration between two solutions.

confidence interval A calculated range in values that estimates where the true value is likely to be, according to a pre-determined level of confidence.

connective tissue A type of tissue that connects, separates or supports tissues or organs in the body.

constant region The region of antibody molecules that remains the same and interacts with receptors on the body's cells.

contact tracing The identification of possible contact with individuals from a source of an infection, determining the number infections, providing care and surveillance of further transmission.

contagious An ability to be spread from one organism to another, generally by direct contact.

contaminant Something that causes an impurity, polluted or a substance to become poisonous.

continuous variable A numerical variable that is based on a continuum in which infinite fractional values exist.

control group The selected test group (sample group) in which no change to the independent variable exists and represents the typical natural conditions.

controlled variable A variable that is controlled during an experiment so that it does not change and does not influence the measured results or dependent variable.

cord blood Blood taken from the umbilical cord after the birth of a baby.

coronary circulation The movement and supply of blood to the heart muscle. Oxygenated blood is supplied to the heart muscle by arteries branching from the base of the aorta. Deoxygenated blood is carried by the veins into the coronary sinus which then enter the right atrium.

cortisol A steroid secreted in small amounts as hydrocortisone from the human adrenal cortex of the adrenal gland generally in response to stress.

countercurrent blood flow A system of heat exchange where blood is warmed or cooled by blood in an adjacent blood vessel flowing in the opposite direction.

Creutzfeldt–Jakob disease (CJD) An infectious disease caused by a misfolded PrP protein.

cyanogenic glycoside A compound produced by plants that breaks down to form hydrogen cyanide, a compound extremely toxic to eukaryotic cells. These compounds assist the plant to resist infection by pathogens and damage by herbivores.

cytokine A chemical used for cell communication, especially between cells of the immune system. Cytokines are peptides or proteins.

cytokinin A plant hormone that, in the presence of auxin, stimulates the division of plant cells.

cytology The study of the structure and function of cells.

cytoplasm The fluid content of a cell, made up mostly of water; includes ions, enzymes, food molecules and organelles other than the nucleus.

cytosol The fluid component of cytoplasm in which organelles are located.

cytotoxic T lymphocyte (T_C) A T lymphocyte that is stimulated by cytokines to bind to antigen MHC-I complexes on the surface of infected host cells and release cytotoxic compounds that destroy the infected cells.

D

DALY (disability adjusted life years) A measure of overall burden of disease. It is a measure of the shortening of life span, which the disease burden causes.

data The measurements or observations collected during an investigation.

data logger A portable tablet that records and logs data acquired or measured by using probes, sensors and instruments.

defensin A protein produced by plants that disrupts the activity of digestive enzymes and which is also thought to damage the cell membranes of microbes. Defensins help to protect plants from infection and production of them increases when the plant is under threat.

denature (or denaturation) An irreversible change in the tertiary structure of a protein (e.g. as a result of heating the protein above a critical temperature).

dendrite A long extension of the cell body that increases the surface area for receiving inputs from other neurons or sensory receptors.

dendritic cell A type of antigen-presenting cell.

dependent variable The variable that is measured or observed during an experiment; is assumed to change due to changes in the independent variable.

diffusion The passive movement of a solute from a region of high concentration to a region of lower concentration.

digestion The breakdown of food into a form that can be used by an organism for metabolism; involves mechanical digestion and chemical digestion.

digestive enzyme An enzyme that assists in the breakdown of otherwise indigestible matter.

digestive enzyme inhibitor An enzyme or lectin that blocks normal digestion of starch by insects.

direct contact transmission The transmission (transference) of a pathogen from one individual to another through physical contact.

discrete variable A numerical variable that is based on defined integer values; fractions between integers do not exist.

disease Any condition that impairs the normal functioning of an organism.

DNA (deoxyribonucleic acid) A double-stranded nucleic acid made up of a sequence of deoxyribose sugars and bases (adenine, cytosine, guanine and thymine) linked by phosphate bonds. It is the carrier of genetic information in all cellular organisms and most viruses; found in chromosomes (and mitochondria and chloroplasts).

dopamine A catecholamine that acts as a neurotransmitter in the central nervous system.

droplet A form of direct contact transmission through a drop of saliva larger than 5 µm ejected by coughing, sneezing, laughing or talking.

Duffy An antigen found on the surface of red blood cells used by the malarial parasite to gain entry to the cell.

E

ectoderm The outermost layer of the three primary germ layers in the early embryo.

ectoparasite A parasite that lives on the outside of its host, usually on the skin or surface.

ectotherm A cold-blooded animal.

effector A muscle or gland that responds to a stimulus.

efferent (motor) neuron A neuron that transmits information from the central nervous system to the tissues and organs (effector cells).

egestion The elimination of food that has not been absorbed by the gut.

eicosanoid A compound derived from the fatty acid arachidonic acid and can act as an intracellular messenger.

electron transfer chain A chain or interconnected series of enzymes and cytochromes embedded in the inner mitochondrial membrane (also called the 'electron transport chain').

embryo The stage in the development of a vertebrate, between the fertilisation of the ovum and the development of mature characteristics (the fetus).

embryonic stem cell A stem cell that can be obtained from blastocysts. Embryonic stem cells are pluripotent and can differentiate into any of the three germ layers (ectoderm, endoderm and mesoderm).

endemic A regularly found or prevalent distribution in a population or area.

endocrine system A system of endocrine glands secreting a variety of hormones.

endocytosis The movement of substances into the cell from the extracellular fluid to the cytoplasm.

endoderm The innermost layer of the three primary germ layers in the early embryo.

endoparasite A parasite that lives within its host.

endoplasmic reticulum A continuous membrane system that forms a series of flattened sacs within the cytoplasm of eukaryotic cells. Endoplasmic reticulum can be 'rough' (associated with ribosomes and is involved in the synthesis of proteins) or 'smooth' (not associated with ribosomes and is involved in the synthesis of lipids).

endotherm A warm-blooded vertebrate.

endotoxin A lipopolysaccharide that is part of the cell wall of gram-positive bacteria.

enzyme A protein molecule that acts as a biological catalyst. Enzymes speed up rates of reactions that would otherwise take place much more slowly. Their action is often specific to only one type of reaction.

enzyme–substrate complex The temporary complex that forms when an enzyme binds to a substrate.

epidemic The sudden increase in incidence of a disease in an area above what is normally expected.

epidemiology The study of the incidence and prevalence of disease and the methods of disease control.

epidermis The outermost layer of cells of a multicellular organism.

epiglottis A thin flap of cartilage that covers the entrance to the larynx, preventing food from entering the trachea while eating.

epithelium A thin layer of tissue covering the external surfaces of a multicellular organism, and also lining the inner surfaces of internal structures such as intestines and lungs.

epitope The part of an antigen that binds to the antigen-binding site of a specific antibody.

ergotism A disease caused by consumption of ergot alkaloids produced by the fungus *Claviceps purpurea,* infecting grains, usually wheat or rye.

error The difference between a measurement and the true value; this is not an indication of human error or a mistake.

ethics The moral principles that govern a person's behaviour or how an activity is conducted.

ethylene A plant hormone associated with fruit development and ripening; also known as ethene.

eukaryotic cell A cell that contains distinct membrane-bound nuclei and many organelles; organisms that are composed of one or more eukaryotic cells are called eukaryotes and include protists, fungi, plants and animals.

excretion The removal of waste substances from the body of an organism.

exhalation Expiration of air from the lungs.

exocytosis The movement of substances out of the cell from the cytoplasm to the extracellular fluid.

exotic incursion The sudden invasion of foreign material or organisms.

exotoxin An extremely poisonous toxin released by bacteria into their surroundings, either directly or during lysis by immune system cells.

experimental group The selected test group (sample group) in which the independent variable has changed to elicit a change in the measured results.

extend To adjust the methodology to address limitations in the scope or applicability of the original data.

external environment The environment immediately surrounding an organism.

extracellular digestion Chemical digestion in which the enzymes are secreted into a cavity where digestion takes place.

extracellular fluid The fluid outside the cells in a multicellular organism.

extrapolate Extend or project information or data beyond what is measured or known to predict or infer an unknown situation.

extremophile An organism that can live in a physically or geochemically extreme environment that most organisms are not able to adapt to.

F

facilitated diffusion The passive diffusion through selective protein channels in membranes.

fact A known and witnessed entity that is distinguishable from transient theories; it is definite, permanent and independent of any subjective interpretation.

$FADH_2$ (flavin adenine dinucleotide) A redox coenzyme that is formed during the Krebs cycle and utilised during the last part of aerobic respiration, the electron transfer chain.

fermentation The stage in the breakdown of glucose that follows glycolysis when there is no oxygen present; produces either lactic acid (in most animals) or alcohol (in most plants and microorganisms).

fetus The stage in the development of a mammal, following the embryonic stage. The fetus has all of the major structures of the adult mammal. In humans, this stage lasts from the eighth week of gestation until birth.

fever An increase in body temperature that results from the set body temperature point being raised by the hypothalamus in response to the presence of inflammatory cytokines. Fever increases the activity of some leukocytes and decreases the activity of some bacteria.

filariform The thin hair-like larvae of nematode worms.

filtration In the kidney, the process by which the primary kidney filtrate is formed; by passing the fluid from Bowman's capsule into the nephron.

fomite An object or material that can carry a pathogen.

fungus (pl. fungi) A non-photosynthetic eukaryote that has a rigid cell wall made of chitin; fungi include moulds, yeasts, mushrooms and toadstools.

G

gall An abnormal growth of plants, similar to a tumour, which is caused by pathogens or insects.

gall bladder Organ that stores and concentrates bile before releasing it to the small intestine.

gastrula An early stage in the development of an embryo, consisting of three layers of cell—ectoderm (outer layer), mesoderm (middle layer) and endoderm (inner layer).

gastrulation A series of cell and tissue movements at the blastocyst stage of animal development, during which the embryo is reorganised to form a gastrula.

gene A section of DNA that contains instructions for making a protein. Particular genes have specific locations on chromosomes. Genes are copied and passed from one generation to the next during reproduction.

genophore A circular DNA chromosome found in prokaryotic cells.

germ layer The primary layer of cells that are formed during embryogenesis. Animals with bilateral symmetry have three layers—endoderm, mesoderm and ectoderm. Animals with radial symmetry have two layers—endoderm and ectoderm.

gibberellin A naturally occurring hormone that accelerates plant growth by increasing stem elongation. Gibberellins are present at active growth sites such as apical buds and root tips.

glomerulus A clump of looping capillaries in the kidney embedded in the Bowman's capsule.

glucose A monosaccharide ($C_6H_{12}O_6$) that is the most common compound from which energy is released in cellular respiration.

glucagon A hormone produced in the pancreas that causes glycogen to be broken down in the liver, releasing glucose into the blood, thus opposing the effect of insulin.

glycogen A complex carbohydrate molecule consisting of glucose subunits; the main carbohydrate storage molecule in animals.

glycolipid A carbohydrate molecule linked to lipids; for example, on the outer surface of a cell membrane.

glycolysis The first stage in the breakdown of glucose to produce ATP; occurs in the cytoplasm and produces a net of two molecules of ATP for each glucose molecule.

glycoprotein A carbohydrate molecule linked to proteins; for example, on the outer surface of a cell membrane.

Golgi apparatus A stack of flattened, smooth membrane sacs, which form vesicles to process and transport the proteins from the cell.

gram-negative Bacteria with a thin layer of peptidoglycan in their cell wall. When stained with crystal violet dye, gram-negative bacteria do not retain the dye and are pink (negative Gram staining result).

gram-positive Bacteria with a thick layer of peptidoglycan in their cell wall. When stained with crystal violet dye, gram-positive bacteria retain the dye and are purple (positive Gram staining result).

Gram stain A stain applied to bacteria that separates bacteria into two groups according to their cell wall chemistry. Cells can either stain gram-positive (violet) or gram-negative (pink).

grana (sing. granum) A stack of thylakoids in the chloroplast.

granzyme A protein that is released by natural killer cells and cytotoxic T lymphocytes, which stimulates body cells to undergo rapid cell death/apoptosis.

guard cell The cells that form a pair of curved cells that surround a stoma, becoming larger or smaller according to the pressure within the cells.

H

haemagglutinin A glycoprotein found on the surface of viruses that assists the virus to attach to the prospective host cell.

haemoglobin A protein molecule in red blood cells that carries oxygen from the lungs, and returns carbon dioxide from the tissues to the lungs; occurs in mammals and many other animals, and gives red blood cells their characteristic colour.

haemolymph Circulatory fluid in the open circulatory systems of invertebrates.

haemolytic disease of the newborn A disease of newborn babies in which their red blood cells are attacked by antibodies that have crossed the placenta from their mother. Caused by Rh incompatibility.

heat exchange The movement of heat between the body of an organism and the surrounding medium, usually air or water.

heavy chain The polypeptide chain that forms the stem of a Y-shaped antibody.

helminth A member of the worm-like phyla; includes nematodes and platyhelminths.

helper T lymphocyte (T_H) A lymphocyte that binds to antigen MHC-II complexes on antigen-presenting cells and then activate B lymphocytes to secrete antibodies, macrophages to phagocytose pathogens and cytotoxic T lymphocytes to kill infected cells.

herbivore An animal that feeds only on producers, such as plants or algae.

herd immunity The phenomenon in which the immunity of a large proportion of a population provides protection from a pathogen to non-immune or non-vaccinated individuals. Often achieved through vaccination and also known as community immunity.

herd immunity threshold The percentage of the population required to have immunity to a pathogen that is estimated to achieve herd immunity.

heterotroph An organism that must obtain nutrients from other organisms.

heterozygote advantage The increase in chances of survival resulting from having a heterozygous genotype.

heterozygous Having two different alleles at the same gene locus on each member of a homologous pair.

hibernation A controlled lowering of the body temperature of an endotherm as a means of reducing energy expenditure. Hibernation reduces the amount of energy required at a time when little food is available.

histamine An organic compound involved in the inflammatory response and allergic reactions. Histamines cause blood vessels to dilate and become more permeable. They also promote fever.

homeostasis The maintenance of a relatively stable internal environment in the face of changes in the internal or external conditions.

homozygous The presence of identical alleles at the same gene locus on each member of a homologous pair.

hormone A substance that is produced by one tissue and transported to another tissue, where it induces a specific physiological response.

human leukocyte antigen (HLA) An alternative name for the major histocompatibility complex (MHC) in humans.

humoral immunity An immune response involving B lymphocytes that produce specific antibodies against specific antigens.

hydrolysis A chemical reaction involving the splitting of a molecule by the addition of a water molecule at a particular point.

hydrolytic enzyme An enzyme, found in plants, that breaks down the cell walls of fungi, oomycetes, bacteria and the exoskeletons of insects. They are produced in greater quantities when the plant is under attack.

hydrophilic The property of an attraction to water, readily mixing with and dissolving in water.

hydrophobic The property of an aversion to water, tending to coalesce and form droplets in water.

hydrophyte A plant that lives in, on or under water.

hygiene The actions that lead to cleanliness and result in good health.

hygiene etiquette The practice, activities, customary code or behaviour of cleanliness, interrupting pathogen transmission and encouraging health.

hypha (pl. hyphae) A long, branching filamentous structure that makes up the bodies of many fungi and oomycetes.

hypothesis A tentative explanation that is based on observation and prior knowledge. A hypothesis must be expressed as a clear statement, and be testable and falsifiable (can be proven false).

I

immune system A system of the body that protects it against pathogens.

immunisation The process of artificially inducing immunity to a particular antigen, usually by administering a vaccine.

immunisation coverage The proportion or percentage of the population that has immunity to a pathogen, achieved through vaccination.

immunogen An antigen that elicits an immune response.

immunoglobulin (Ig) A type of protein, produced by B lymphocytes in an immune response to a specific antigen; also called an antibody.

immunosuppressed The suppressed state of an individual's immune system, resulting in slowed adaptive immune responses to pathogens. Immunosuppressed individuals are at higher risk of disease due to the slow immune system response.

impermeable membrane A membrane that does not allow substances to pass through it.

inactivated vaccine A vaccine made from inactivated (often killed) forms of a pathogen. Inactivation destroys the pathogen's ability to replicate and cause disease, but its antigens are preserved so that they can be recognised by the immune system upon future exposure.

incubation period The period of time from infection until symptoms appear.

independent variable The variable during an experiment that is deliberately altered, or selected to be tested, which is assumed to cause a change in the dependent variable.

indirect contact transmission The transmission (transference) of a pathogen from an infected individual to another through non-physical contact.

infectious A pathogenic molecule or organism that can be spread from one organism to another organism.

infectious disease A medical condition or disease that is caused by infectious agents (pathogens). Infectious diseases are transmissible.

inference A conclusion derived from evidence and reasoning without witnessing every explicit detail; made by listening or reading beyond what has been literally expressed; an implication.

inflammation A protective response triggered by damaged tissues or invading pathogens, which leads to increased blood flow to the area of damage/infection, along with migration of leukocytes to those tissues. It results in heat, pain, swelling, redness and loss of function.

inhalation The physical action of inhaling or breathing in.

innate immune response An immune response that non-specifically protects against a wide variety of pathogens. It consists of physical, chemical and microbiological barriers that provide resistance to infection, and an innate response to infection that involves leukocytes such as phagocytes and defensive molecules such as complement proteins.

inorganic compound A compound that lacks a carbon atom.

insulin A hormone in vertebrates secreted by the beta cells of the pancreas, controlling the concentration of glucose in the blood.

integral protein A protein molecule that is a permanent part of the cell.

interferon A cytokine that is a signalling molecule important in animal immunity. Interferons are produced by virus-infected cells to inhibit viral replication and strengthen the barriers of surrounding cells. They interfere with the transcription of viral genes and also regulate the immune response by enhancing the activity of T lymphocytes.

interleukin A type of cytokine produced by leukocytes for regulating immune responses.

intermediate host The host in the life cycle of a parasite in which larval forms grow and develop.

internal environment The environment within an organism.

interneuron A neuron that connects neurons within the nervous system.

interstitial space The space between cells.

intracellular digestion The breakdown of particles that takes place in the cytoplasm of a cell.

intracellular fluid The fluid contained within the cell membrane; the fluid of cytoplasm.

K

karyotype A picture of an organism's full set of chromosomes, arranged in homologous pairs.

killed vaccine A particular type of inactivated vaccine where the pathogen, such as a bacterium, used in the vaccine has been killed.

kleptothermy A form of thermoregulation in which animals (either voluntarily or involuntarily) share body heat.

Krebs cycle The second stage of aerobic cellular respiration, the oxidative metabolism of acetyl units to produce high-energy compounds such as ATP. From each turn of the Krebs cycle, one molecule of acetyl CoA is metabolised into two molecules of carbon dioxide, three molecules of NADH, one molecule of $FADH_2$ and one molecule of GTP (used to make one ATP).

L

lacteal The vessels of the lymphatic system close to the small intestine. Lacteal capillaries absorb digested fats from the small intestine, giving the lacteals a milky appearance.

lamellae (sing. lamella) Thin layer or membrane; membranous fold in a chloroplast.

larynx The organ in the upper trachea that contains vocal cords and is responsible for speech in humans. It is also involved in breathing and in preventing the aspiration of food or other large particles into the lungs.

law A causal relationship that describes or predicts phenomena under known conditions; the known cause of any effect in a system where the variables and conditions are known.

lenticel A porous group of cells that allows gas exchange across the otherwise airtight and waterproof cork layer covering the stems and roots of woody plants.

leukocyte A white blood cell; including phagocytes and all lymphocytes.

light chain The short polypeptide chain that forms the arms of the Y-shape.

lignin A complex organic polymer deposited in the cell walls of many plants, making them rigid and woody.

line graph A graph that displays continuous data by a line from one point of measurement to the next.

linear relationship A direct relationship between two variables in which a change in one is proportional to the change in the other.

lipase An enzyme that hydrolyses lipids.

lipid An organic compound that is insoluble in water but soluble in alcohol, ether or chloroform. Lipids include fats, oils, sterols, some hormones, fat-soluble vitamins, glycerides and phospholipids.

literature review An evaluative report that critically analyses the current body of knowledge.

live attenuated vaccine A vaccine that uses a living but weakened form of a pathogen in order to stimulate an immune response. The weakening of the pathogen ensures that it cannot cause disease.

liver A large organ in vertebrates that is involved in many important metabolic processes, including protein manufacturing, fat storage and processing, bile secretion and metabolism of toxins.

loop of Henle A U-shaped loop in a mammalian kidney between the proximal and distal convoluted tubules, dipping into the medulla. Its main function is to recover water and sodium chloride from urine, thus making the urine more concentrated and reducing the amount of water that needs to be taken in.

lumen (1) The region enclosed by the cell membrane of a cell. (2) The inside of a tubular structure such as a blood vessel or xylem tube.

lymph The fluid that circulates in the lymph system. It consists mainly of interstitial fluid (fluid forced from capillaries by blood pressure into the spaces between tissues) and contains lymphocytes, macrophages, proteins and fats. It has an important role in defending the body against harmful bacteria and other particles, and also in the absorption and transport of fatty acids.

lymphocyte A white blood cell formed or matured in lymphatic tissue such as B cells, T cells and natural killer cells. B cells and T cells are fundamental to the adaptive immune response.

lysosome A specialised vesicle that digests unwanted matter.

M

macrophage A type of leukocyte that can engulf non-self material by phagocytosis.

major histocompatibility complex (MHC) A group of major histocompatibility proteins found on the surfaces of cells, which are involved in antigen presentation. MHC proteins are also known as human leukocyte antigens.

malnutrition A deficiency or excess of nutrients, protein or energy that has an adverse effect on the body.

mast cell An immune cell found embedded in many tissues, especially connective tissues, which releases histamine to induce inflammation when activated or damaged. Mast cells are strongly involved in allergic reactions such as hay fever and anaphylaxis.

mathematical modelling The development of a model that describes a system, concept or idea using mathematical language.

mean A calculated value that represents the centre of a set of numbers that have been averaged.

measure of central tendency A value that estimates the central position in a set of data.

measured variable A variable that cannot be controlled and so must be measured during an experiment to determine whether it influences the measured results or dependent variable.

measurement uncertainty The uncertainty in a measurement due to the precision of the measurement instrument. For an analog instrument it is usually half the smallest increment. For a digital instrument it is the smallest increment on the readout.

mechanoreceptor A sensory receptor that detects and responds to a change in shape.

median The value in the middle of an ordered list of values.

megakaryocyte A large cell in the bone marrow that forms platelets.

memory T lymphocyte A type of T lymphocyte which remains in the body, usually in lymphoid tissues, after an infection. They can be activated upon subsequent infections with the same pathogen to provide a faster and larger response than the first infection.

merozoite An amoeba-like spore produced by certain protozoan parasites; including the parasite that causes malaria.

mesoderm The middle layer of the three primary germ layers in the early embryo. The mesoderm is present only in animals with bilateral symmetry; animals with radial symmetry have only two primary germ layers.

mesophyte A terrestrial plant living under moist conditions with well-aerated soils.

metabolism The total of the physical and chemical processes by which energy and matter are made available by an organism for its own use; is controlled by enzymes.

methodology A systematic, structured approach to conducting a scientific experiment or investigation, outlining the step-by-step procedures used to gather data and apply the scientific method.

microbe-associated molecular pattern (MAMP) A specific molecule typical of certain pathogens found on the surface of their cell wall or cell membrane that can be recognised by receptors found in plant cells and which cause an immune response in the plant. Each subsequent infection by the same pathogen elicits the same response.

microvilli (sing. microvillus) A microscopic fold of the inner surface of intestinal epithelial cells. Microvilli increase the surface area for the absorption and secretion of substances.

mitochondria (sing. mitochondrion) Organelle responsible for energy transformation within the cell.

mitosis A division of a nucleus that results in two cells that are genetically identical to the parent cell. Asexual reproduction and cell replication for growth occur by mitosis.

mode The value that occurs most often in a list of values.

model A representation of a structure or process, such as a physical model or a digital model, that is used to create and test theories and explain concepts.

modify To change the form or qualities of something.

morbidity The physical state or condition of being diseased.

morula An embryo consisting of an unorganised mass of 16 cells, resulting from a series of divisions of the zygote.

mRNA (messenger RNA) An RNA molecule transcribed from DNA in the nucleus, which passes into the cytoplasm and binds to a ribosome, where it is translated into an amino acid sequence (polypeptide).

mucous membrane A lining or thin tissue of the body that separates the internal environment from the external and secretes a protective mucus. Mucous membranes line many body cavities and tubular organs, including the gut and respiratory passages, as well as the eyes and nose.

multicellular Consisting of two or more specialised cells that have identical DNA, are responsible for specific functions, and depend on each other for survival. Also called pluricellular.

multipotent A cell that can develop only into cells of a similar type. For example, stem cells in bone marrow are multipotent because they can develop into different blood cells but not into other types of cells.

murein A polymer consisting of sugar and amino acids which form the cell wall of most bacteria.

mutation A permanent change in the genetic sequences of an organism, including changes in the nucleotide sequences or chromosomal arrangements.

myelin sheath The fatty sheath surrounding nerves.

myoglobin A red respiratory pigment that occurs in muscles; carries oxygen that can be used when other oxygen reserves are depleted.

N

NADH (nicotinamide adenine dinucleotide) A similar compound to $FADH_2$ but delivers more H^+ and electrons to the electron transfer chain as well.

natural active immunity The active immunity formed as a result of survival from a natural infection with a particular pathogen.

natural passive immunity The transfer of antibodies from one individual to another by natural means; the transfer of antibodies from mother to fetus via the placenta during fetal development and from mother to baby in breast milk.

negative feedback loop A mechanism in homeostasis whereby a change in the physiological condition triggers a response that re-establishes homeostasis.

nematode A worm of the phylum Nematoda, including hookworms, roundworms and threadworms. Many nematodes are parasitic.

nephron The functional unit of the kidney; consisting of a Bowman's capsule surrounding a glomerulus and a tubular region leading into a collecting duct. About one million nephrons are found in each human kidney.

nervous system System that generates and transmits signals throughout an animal's body, coordinating movement, basic bodily functions and sensory information.

neuraminidase An enzyme produced by viruses, especially those that escape by budding, which facilitates its release from the host cell.

neuron A nerve cell, including its body, dendrites and axon, forming the fundamental unit of the nervous system in animals.

neurotransmitter A chemical released from a neuron ending in response to a nerve impulse that interacts specifically with receptors on a responding cell.

nitrogenous waste The waste products from the breakdown of proteins, including ammonia, urea and uric acid.

nociceptor A receptor sensitive to painful or injurious stimuli.

node of Ranvier A constriction of the myelin sheath occurring at intervals along myelinated nerve fibres, and at which the axon membrane is exposed.

nominal variable A categorical variable in which there is no inherent order. Nominal variables can be counted but not ordered.

non-infectious disease A medical condition or disease that is not caused by infectious agents (pathogens). Non-infectious diseases are non-transmissible.

non-shivering thermogenesis The increased production of metabolic heat by oxidation of fat.

nucleolus A dark-staining body in the nucleus; site of synthesis of ribosomal RNA.

nucleotide Monomer, or building block, of the nucleic acids DNA and RNA. Consists of a phosphate, a sugar and a nitrogenous base.

O

observation Witnessing an event or phenomenon.

omnivore An organism that feeds on both plants and animals.

oomycete A fungus-like pathogen with branching hyphae, called haustoria, that penetrate living tissue and absorb nutrients, or release enzymes that digest cytoplasm into molecules that can be absorbed.

open circulatory system A system for fluid circulation in animals in which there is no clear distinction between circulatory and interstitial fluids, so that fluids flow more or less freely between the cells of the tissues.

ordinal variable Categorical variables in which the order of the nominated category is important.

organ A structure, consisting of different tissues, that carries out one or more specific functions.

organelle A structure in eukaryotic cells that is surrounded by, or consists of, membranes (e.g. nucleus and mitochondria).

organic compound A complex molecule containing carbon and hydrogen, which occurs in living organisms (e.g. proteins, carbohydrates and lipids).

organism A living system that functions as an individual, whether unicellular or multicellular.

osmolality A measure of the concentration of particles (e.g. sodium and chloride ions) that affect osmosis.

osmoreceptor A receptor that is sensitive to blood solute concentrations.

osmosis The passive diffusion of free water molecules across a selectively permeable membrane from a solution in which there are more free water molecules (a dilute solution) to a solution in which there are fewer free water molecules (a concentrated solution).

osmotic gradient The difference in the concentration between two solutions on either side of a semipermeable membrane.

osmotic pressure The pressure causing water to move along a concentration gradient.

outlier A data point that lies outside the main group of data.

oxygen (O_2) Chemical element with the symbol O. Oxygen is an essential element in the biological processes of photosynthesis and cellular respiration.

oxygen-carrying capacity The amount of oxygen that can be carried by a particular medium, such as blood.

oxyhaemoglobin A bright red substance formed by the combination of haemoglobin with oxygen, present in oxygenated blood.

oxytocin A hormone secreted by the posterior pituitary gland, which causes the uterine wall to contract during birth and releases milk from the mammary glands within the breast.

P

pandemic An epidemic that has spread over a large geographical area. It usually affects large numbers of people and may be worldwide.

parasite An organism that lives in or on another organism, called the host, and derives its nutrients from the host to the detriment of the host.

partial pressure The pressure a particular gas contributes to the total pressure of a gas mixture. The partial pressure of gas X is written as P_X.

passive immunity The temporary acquisition of immunity to a particular pathogen through the transfer of antibodies from one organism to another. This may be artificial through injection of antibodies or natural as in the transfer from mother to child.

passive transport The diffusion of molecules across the cell membrane without the expenditure of energy.

pathogen An infectious agent that can induce disease; can be cellular (e.g. bacteria, fungi, helminths) or non-cellular (e.g. prions, viruses, viroids).

pathogen-associated molecular pattern (PAMP) A specific molecule typical of certain pathogens found on the surface of their cell wall or cell membrane, which can be recognised by receptors found in animal cells and cause an immune response in the animal. Each subsequent infection by the same pathogen elicits the same response.

pathogenesis The origin, development and effects of a disease.

pathogenicity The ability of a parasite to cause disease in a host.

pattern recognition receptor (PRR) A receptor on the surface of cells that recognises typical pathogen-associated molecules; MAMPs or PAMPs.

peptidoglycan A macromolecule made of protein and carbohydrate, found only in bacteria.

Pearson's correlation coefficient A calculated estimate of the strength between variables for a relationship of linear regression, producing a line of best fit.

perforin A chemical produced by certain cells, such as natural killer cells, which causes perforations in the cell membrane of cells infected by viruses or which are abnormal, thereby killing the cell.

peripheral nervous system (PNS) The nerves outside of the brain and spinal cord.

peripheral protein A protein that binds to integral proteins or penetrates only one surface of the cell membrane.

peristalsis The coordinated muscular contractions and relaxations of the wall of the digestive tract that move a bolus of food from the oesophagus to the intestines.

personal protective equipment (PPE) The equipment specifically designed and used during activities to protect the user from hazards.

phagocyte Any cell capable of engulfing or phagocytosing pathogens or foreign particles.

phagocytosis The entry of large particles such as bacteria and cell debris into the cell.

phagolysosome A structure formed when a lysosome fuses with a phagosome so that the pathogen or foreign particle can be digested.

phagosome A vesicle formed around a pathogen or foreign particle in order to phagocytose it.

pharynx The tube or cavity, with its surrounding membrane and muscles, that connects the mouth and nasal passages with the oesophagus.

phenolic A protective chemical found in plants, which includes tannins and phytoalexins. Phenolics bind to salivary proteins and enzymes thereby de-activating them. They cause the death of the pathogen through inadequate energy intake.

phenomenon (pl. phenomena) An event that can be observed, measured and described.

phloem The plant tissue through which sugars and other organic compounds are distributed to different parts of a plant. In flowering plants, phloem consists of sieve tubes, companion cells and fibres.

phospholipid A fat-like substance, usually based on glycerol, that is an essential component of cells and is involved in the uptake of fats and fatty acids from the products of digestion.

photoreceptor A sensory receptor that detects and responds to light.

photosynthesis The process by which plants and other photosynthetic organisms convert energy from sunlight into chemical energy for biological function. It occurs in plastids.

phytohormone A plant hormone, including auxins, gibberellins, cytokinins and abscisic acid.

pie chart A spherical display of either qualitative or quantitative data showing the proportion of each data set (measurement, independent variable, observation) as a part of the whole.

pili (sing. pilus) A hair-like structure found on the surface of bacteria that they often use to adhere to a host tissue or cell.

piloerection The innervation of muscles to hair follicles, causing them to rise (goose pimples).

pinocytosis The entry into cells of extracellular fluid and substances such as proteins and sugars that are dissolved in the fluid.

plaque A protein aggregation caused by prions.

plasma cell An activated B lymphocyte that produces large numbers of the same type of antibody.

plasmid A small ring of double-stranded DNA.

plasmodesmata A microscopic channel that connects the cytoplasm of adjacent cells in plants and some algae.

plasmodium A member of a genus of parasitic protozoans; includes the parasites that cause malaria.

platelet A cell fragment, lacking nuclei, found in the blood in large numbers; assist with blood clotting.

platyhelminth A worm that includes flatworms, like tapeworms, and flukes such as the liver fluke.

pluripotent A cell that can develop into several different cell types. An example is a human embryonic stem cell, which can form all adult cell types.

polarised (1) The property of having a charge across a surface. (2) Restricting light to a single plane.

positive feedback loop A type of control mechanism in which the end-product of a particular pathway or process activates the first step in the pathway or in other pathways that contribute to the overall process.

precipitation The formation of a solid substance when two solutions are combined.

precision The ability to consistently obtain the same measurement. To obtain precise results, you must minimise random errors.

primary data Data from an investigation that you have conducted yourself (also called first-hand data).

primary host The host in the life cycle of a parasite in which adult forms produce gametes that fuse to form the larval forms.

primary immune response The immune response to an antigen when it is being encountered for the first time.

primary source The original source from which information was created or data was measured.

principle The known cause of any effect, in a system where the variables and conditions are known.

prion A small protein particle that, when its shape is altered by mutation, causes protein aggregation and is toxic to neurons.

processed data Data that has been manipulated to produce meaningful information; a set of displayed data in an appropriate form such as tables or graphs; required information that has been extracted from a set.

prokaryotic cell A cell that lacks a membrane-enclosed nucleus and membrane-enclosed organelles; organisms that are composed of prokaryotic cells are called prokaryotes and include bacteria and archaea.

prostaglandin A hormone-like signalling molecule used by the body. Prostaglandins are involved in muscle contractions during birth, control of blood pressure, and mediating fever and inflammation.

protease An enzyme that digests proteins.

protease inhibitor A chemical produced by plants to protect against pathogens and herbivores which blocks the activity of proteases used for digestion by the invader.

protein An organic compound composed of long chains of amino acids; also called a polypeptide. These compounds perform structural, contractile, transport, catalytic, nutritional, structural, some hormonal and other functions.

proteomics The study of the structure and function of proteins in organisms.

protist Eukaryotic organism belonging to the kingdom Protista. Protists are mostly unicellular organisms and include protozoa, slime moulds, water moulds and many algae.

psyllid A lice-type insect that feeds on plant tissue and vascular fluids and which may stimulate the formation of galls.

pulmonary artery Artery that carries blood from the right ventricle of the heart to the lungs for oxygenation.

pulmonary vein A blood vessel that in humans carries oxygen-rich blood from the lungs to the left atrium of the heart.

pyruvate A three-carbon molecule formed by splitting a glucose molecule during glycolysis. In aerobic respiration, pyruvate is transported into the mitochondria by active transport where it is oxidised to a two-carbon acetyl group that becomes part of the Kreb's cycle.

Q

qualitative data Non-numerical data that is often descriptive in nature.

quantitative data Numerical data.

quarantine The isolation of an infected person or animal in a place, state and/or time to prevent transmission of disease.

R

random error An error that affects the measurement in an unpredictable way, resulting in an even variation (fluctuation) in measurements above and below the expected true value.

random selection The process of using a selection process that is not controlled by the experimenter.

range The variation in values between upper and lower limits on a particular scale or the largest and smallest measurements.

raw data The data recorded during an experiment that has not been analysed.

reabsorption A process in the kidney, in which the non-waste substances in the primary kidney filtrate are taken back into the tissues via nephrons.

receptor A specialised structure that can detect a specific stimulus and initiate an action potential.

recombinant DNA technology A technique that combines DNA from two or more sources in cell cultures to create a new DNA molecule or genetic sequence.

redirect To adjust the methodology to explore new aspects or gain deeper insight into the phenomena observed in the initial experiment.

reference list A list of resources that are cited or referred to in the document.

refine To adjust the methodology for greater accuracy or precision, often leading to more investigable research questions.

reflex A rapid, unconscious response.

regression The calculated estimate of the strength of a relationship between variables.

regulatory T lymphocyte A T lymphocyte that downregulates the immune response after an infection has been resolved.

relative uncertainty The measured uncertainty displayed as a percentage of the measurement.

reliability The ability to reproduce experimental results under the same conditions.

renin An enzyme released from kidneys in response to changes in blood osmolality or pressure responses.

repeat trial The process of repeatedly conducting a process, activity or trial in an attempt to replicate the results.

replication The process of repeatedly conducting or choosing a process, activity or sample in an attempt to replicate the results, event or set-up.

reproduction number (R_0) The number of infections transmitted from a single case of infection; the number of secondary infections.

research question A focused, specific and relevant query developed from a claim that directs the scientific investigation. It should address a key concept related to the claim and guide all stages of research, including inquiry, analysis, interpretation and evaluation.

reservoir A biological entity that allows infestation of a pathogen through reproduction or multiplication, becoming a source of further infections.

retrovirus A type of virus that uses RNA as its nucleic acid. It uses the enzyme reverse transcriptase to copy its RNA into DNA as one of the first steps in infecting a host cell.

reverse transcriptase A type of polymerase enzyme, used by retroviruses to copy their RNA into DNA as part of the processes of viral infection of a host cell.

rhabditiform The first larval stage of *Strongyloides*, which can develop either into a free-living sexually reproducing adult or into infective filarial form larvae.

ribosome A tiny organelle often attached to the endoplasmic reticulum; composed of protein and RNA; the site of protein synthesis.

risk assessment A systematic process of evaluating and identifying the potential risks associated with an activity and control measures to reduce those risks.

RNA (ribonucleic acid) A nucleic acid made up of a sequence of ribose sugars and bases (adenine, cytosine, guanine and uracil) linked by phosphate bonds.

root hair A very thin extension of an epidermal cell of a root. Root hairs increase the root's surface area, making the absorption of water and minerals from soil more efficient.

root pressure Osmotic uptake of water that accompanies the active uptake of mineral salts and contributes to the movement of water up xylem in some plants.

rough endoplasmic reticulum Layers of intracellular membranes associated with ribosomes. Rough endoplasmic reticulum is involved in protein synthesis.

rRNA (ribosomal RNA) An RNA molecule synthesised in the nucleolus; it forms part of ribosomes.

rumen A large fermentation chamber for the digestion of cellulose, located before the stomach in many herbivorous animals, such as cattle and sheep.

S

safety data sheet (SDS) A document that outlines specific details about items that are required to be considered when handling the item for experimentation.

sample size The number of individual measurements or observations undertaken for each tested variable.

sanitation The provision and conditions, facilities and infrastructure resulting in cleanliness and health.

saponin A soap-like compound present in plants that breaks down lipids and that disrupts the cell membranes of potential pathogens.

scatterplot A graph that displays the measured values for two variables plotted along two axes.

scientific journal A book containing the entire collection of work related to an experiment or investigation.

scientific method An orderly process of determining the relatedness between phenomena that were originally witnessed and caused a query to develop.

scientific notation A measured value written as a mathematical expression, between 1 and 10 including decimals, displaying all significant figures, multiplied by ten raised to a specific power.

scientific report A document that follows the report genre, systemically organising the main information and findings of scientific research or work.

secondary data Data or information you have not collected or produced yourself (also called second-hand data).

secondary immune response The immune response to an antigen that was previously encountered and provoked an immune response. The process activates memory cells and so is faster and stronger than the primary response.

secondary infection The infection that occurs after the initial infection or case. May be due to the primary or initial infection.

secondary source A source of information or data that was not created or measured by its author, but which cites another source.

secretion (1) The release of specific substances from a cell or group of cells. (2) In kidneys, the active excretion of particular substances by the cells of the duct walls.

selection bias An inclination or prejudice towards something during the process of selecting samples.

self-tolerance The inability of the immune system to respond to self-antigens.

semipermeable membrane A membrane that allows only some molecules to pass across it by osmosis or diffusion. Also called partially permeable membrane or selectively permeable membrane.

sensory neuron A type of nerve cell that transmits nerve impulses from sensory receptors to the central nervous system.

serum The fluid portion of the blood that remains once the blood cells and clotting factors have been removed.

shivering thermogenesis The increased production of metabolic heat through shivering.

signal transduction The passage of signals along nerve axons.

significant figures The digits recorded according to the precision of an instrument; the digits in a calculation that represent the precision of an instrument.

sink A site where something is stored or consumed.

smooth endoplasmic reticulum A continuous membrane system that forms a series of flattened sacs within the cytoplasm of eukaryotic cells. It is not associated with ribosomes and is involved in the synthesis of lipids.

solute A substance that is dissolved in a solution.

solvent The dissolving agent of a solution, usually water.

soma (1) The body of an organism with the exception of its reproductive cells (gametes). (2) The central part of a neuron (nerve cell) that contains the nucleus; also called a cell body.

source (1) A site where something is produced. (2) A document or person from which information has been obtained.

Spearman's rank correlation A calculated estimate of the strength between variables for a non-linear relationship, producing a line of best fit.

specialised cell A cell that has a specific function (e.g. a red blood cell).

standard deviation A calculated spread of data that estimates the quartile boundaries from the mean, assuming a normalised (bell) curve.

starch A complex carbohydrate consisting of glucose subunits; the main form of energy storage in plants.

stem cell A cell that can differentiate into a specialised cell.

steroid A polycyclic lipid with a hydrocarbon nucleus; steroids include bile acids, sterols and various hormones.

stimulus An agent that causes a reaction or change in an organism or in any of its parts.

stomata (sing. stoma) A pore in the leaf epidermis, bounded by specialised guard cells that open and close the pore. Stomata are the main routes through which gas exchange occurs in plants, and through which water loss is regulated.

stroma (pl. stromata) The fluid matrix part of a chloroplast in which the light-independent reactions occur.

Student's *t*-test A calculation that produces a value that estimates the likelihood that two variables are not related.

substrate A molecule that is acted on by an enzyme.

subunit vaccine A vaccine that contains only antigens from a pathogen. One or more antigens may be included to stimulate an immune response.

susceptible The state of being open to influence or an agent of influence; likely to be influenced by a particular agent.

symporter A protein carrier molecule found in the cell membrane that transports two different molecules across the membrane in the same direction.

symptomatic An organism that displays symptoms while infected and/or diseased.

synapse The point of communication between two cells, where at least one of the cells is a neuron. It includes the membrane of the presynaptic neuron, the synaptic gap and the membrane of the postsynaptic cell (which may be, for example, a neuron, muscle cell or gland cell).

synaptic terminal The point of communication between one nerve cell and another, or a nerve cell and a target cell such as a muscle or gland.

systematic error An error in measurements caused by the design of a system (e.g. methodology) or instrument (without calibration) that results in the measurements shifting in a systemic direction.

T

T cell receptor (TCR) A receptor on the surface of T lymphocytes that binds to antigens displayed by antigen-presenting cells attached to MHC-II proteins. This binding activates the T lymphocyte and provides the link between the innate and adaptive immune responses. It is made up of two polypeptide chains that have a variable and a constant region and only one antigen-binding site.

T lymphocyte (or T cell) A type of lymphocyte that originates in the bone marrow and matures in the thymus gland. It is responsible for cell-mediated immunity. T lymphocytes include cytotoxic T lymphocytes, helper T lymphocytes, regulatory T lymphocytes and memory T lymphocytes.

taxonomy The system of naming groups recognised in a classification of organisms.

terpene A chemical produced by plants that is highly toxic to fungi and many insects. In insects, terpenes mimic certain hormones, causing disruptions to their life cycle. Pyrethrins and phytoectysones are included in this group.

theory A set of concepts, claims or laws that explain and predict phenomena. A theory is supported by a vast body of evidence that is reproducible and is from a wide variety of sources.

thermoreceptor A sensory receptor that responds to change in temperature.

thermoscanning Using a device to produce a thermal image by detecting infrared radiation, showing the temperature of the surface of an object and the variations in the surface temperature.

thylakoid A membrane-bound compartment inside a chloroplast. Thylakoids are the site of light-dependent reactions in photosynthesis.

thyroid hormone A hormone secreted from the thyroid glands which regulates the rate of metabolism in the body.

tidal volume The volume of air moved into and out of the lungs during breathing.

tissue A group of similar cells functioning together.

tonoplast A large permanent vacuole surrounded by a membrane in plant cells

torpor A state of dormancy for a brief or long period, during which the metabolic rate is slow and the animal becomes unresponsive to external touch or noise.

totipotent A cell that can give rise to any cell type and potentially a completely new organism.

toxin A naturally occurring poison produced by living cells or organisms, such as bacteria, fungi, plants or animals.

toxoid vaccine A type of subunit vaccine that uses toxins, inactivated by the chemical formalin, to elicit an adaptive immune response.

trachea The tube in humans and other air breathing vertebrates extending from the larynx to the bronchi, serving as the principal passage for conveying air to and from the lungs; the windpipe.

tracheid A long, hollow cell with a thickened wall and tapering ends, found in the xylem of vascular plants. Tracheids transport water and nutrients to the living cells of the plant.

translocation The transport of organic substances in the phloem of a vascular plant.

transmembrane protein A type of membrane protein that spans the entire width of the membrane; it is often involved in transport of substances across the membrane.

transpiration The loss of water from the leaves of plants through stomata. Transpiration causes suction, which draws water up xylem vessels from the roots.

transpiration stream The flow of water within a plant, from the uptake by the roots to the loss of water to the environment via the leaves.

trend line A line drawn onto a graph to accurately display a relationship between the variables.

trichinosis A disease caused by the ingestion of the parasite *Trichinella spiralis*, usually in raw or undercooked meat.

true value An ideal measurement of a phenomenon without error.

turgor A state of high internal fluid pressure that is the result of the osmotic intake of water into plant cells whose volume is limited by the rigid cell wall.

U

uncertainty A range of measurements from the experiment within which the true value is thought to be.

uncertainty of the mean The uncertainty associated with the average of several measurements. It is half the range of the set of measurements.

unicellular An organism consisting of a single cell.

uniporter A protein carrier molecule found in the cell membrane that transports one molecule across the membrane in one direction.

unipotent A stem cell that can differentiate only into one cell type.

urea A water-soluble diamide molecule (CH_4N_2O) that is a major product of protein breakdown. It is excreted by many vertebrates, including mammals.

ureter The tube that carries urine from a kidney to the bladder for storage, before release of urine via the urethra.

urethra The tube that carries urine from the bladder to the exterior for excretion.

uric acid A complex nitrogenous compound ($C_5H_4N_4O_3$) that is produced and excreted by birds and most land reptiles.

V

vaccination The process of introducing a vaccine into the body in order to elicit an adaptive immune response and create memory cells so future responses will be quicker and stronger if the antigen is encountered in the future. The process of creating artificial active immunity.

vaccine A preparation of altered, weakened or killed microorganisms, or inactivated forms of toxins or antigens introduced into the body (usually by injection) in order to elicit an adaptive immune response.

vacuole An organelle involved in storage in plant cells.

validity A measurement is said to be valid if it measures what it is supposed to be measuring. An experiment is said to be valid if it investigates what it sets out and/or claims to investigate.

valve A specialised structure in closed circulatory systems that allows movement in one direction only. In humans, valves occur in the heart, veins and lymph vessels.

variable A factor or condition that can change during an experiment or investigation.

variable region The region of an antibody that varies between different antibodies and allows them to interact with different antigens.

vascular bundle A grouping of vascular tissues in vascular plants, containing both xylem and phloem. Vascular bundles are continuous, from the roots into the stem, branches and leaves.

vascular plant A plant that has vascular tissues (xylem and phloem) in which the cell walls contain lignin. All living plants, except bryophytes, are vascular plants.

vascular tissue The tissue that conducts water and nutrients around vascular plants. It consists of two types of tissue—xylem and phloem. Vascular tissue also provides structural support to a plant.

vascularised tissue The tissue that contains many blood vessels.

vasoconstriction The constriction of a blood vessel with a reduction of the lumen as a result of contraction of the smooth muscle in the blood vessel wall.

vasodilation The enlargement of the calibre of blood vessel by relaxation of smooth muscle in the blood vessel walls.

vector (1) An infectious disease. (2) An object or organism that transfers a parasite from one organism to another.

vein A blood vessel that carries blood towards the heart. All veins except the pulmonary veins carry deoxygenated blood.

ventilation The active movement of air or water past gas exchange surfaces in animals. In land animals, it is called breathing.

ventricle A muscular chamber of the heart that pumps blood out of the heart. In a four-chambered heart (as in humans) the right ventricle pumps deoxygenated blood to the lungs, and the left ventricle pumps oxygenated blood to other body tissues.

venule A small vessel that connects capillaries to a vein.

vesicle A membrane-bound organelle often involved in transport within the cell.

villi (sing. villus) A tiny fold in the lining of the intestine. Villi increase the surface area available for the absorption of food.

virion A single virus particle.

viroid Infectious agent of plants that is a type of self-cleaving RNA enzyme (or ribozyme); composed of short, circular stands of RNA that lack a protein coat.

virulence A measure of the degree of pathogenicity of a particular pathogen (i.e. the degree to which it causes disease).

virus An infectious agent composed of genetic material (either DNA or RNA) enclosed in a protein coat, and sometimes also a lipoprotein envelope, which is only able to multiply in a host cell.

vital capacity The maximum volume of air that can be moved into and out of the lungs in one breath.

X

xerophyte A plant that has adaptations that conserve moisture and prevent the leaf temperature from rising too much.

xylem The tissue in vascular plants that transports water and nutrients upwards from the roots; it consists of hollow chains of dead cells.

xylem vessel A long tube consisting of cells joined end to end, through which water and nutrients are transported from the roots to the leaves in a vascular plant.

Z

zoonotic disease An infectious disease that passes from non-human animals to humans or from humans to non-human animals.

zoospore An asexually reproduced reproductive cell of fungi, oomycetes, protozoans or algae that possesses flagella and is thus motile.

zygote Diploid cell resulting from the fusion of an egg and sperm. The zygote is the first stage of animal development after fertilisation and before cell differentiation.

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